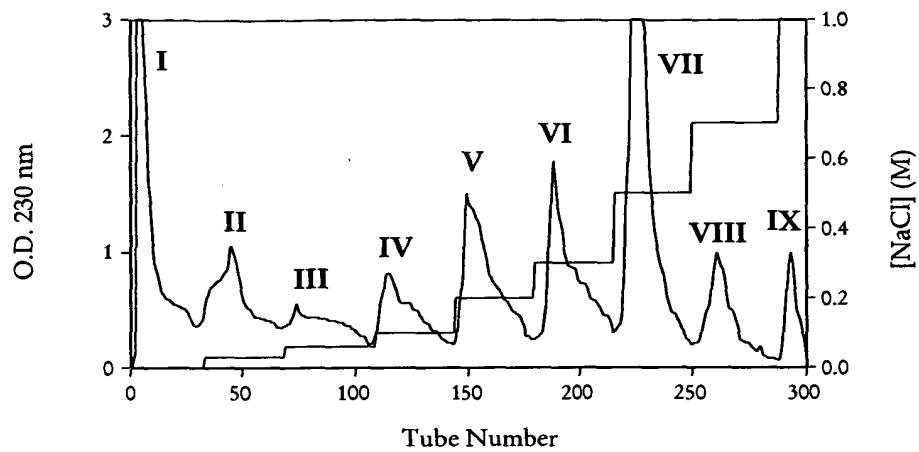


Fig. 1.0

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A.



B.

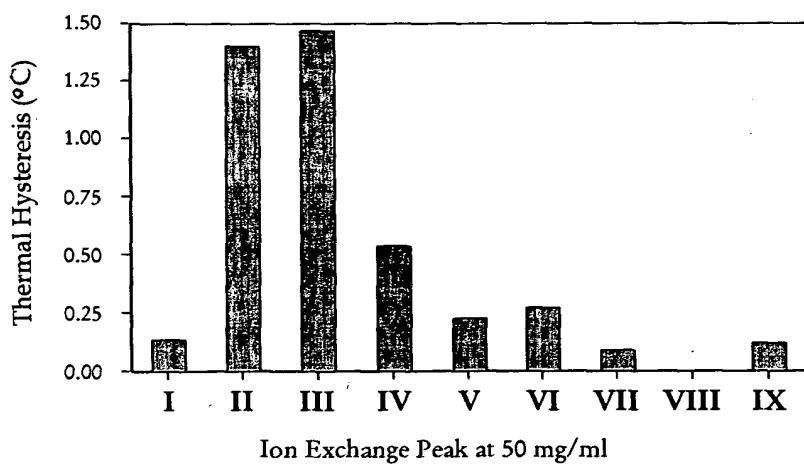


Fig. 1.1

2023-10-29 22:50

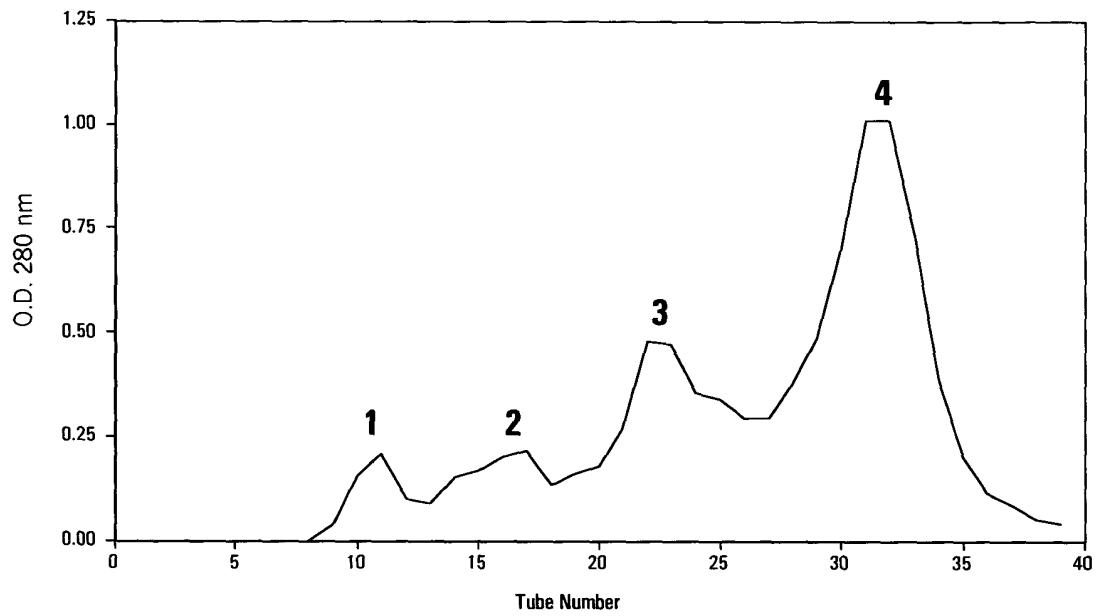


Fig. 1.2

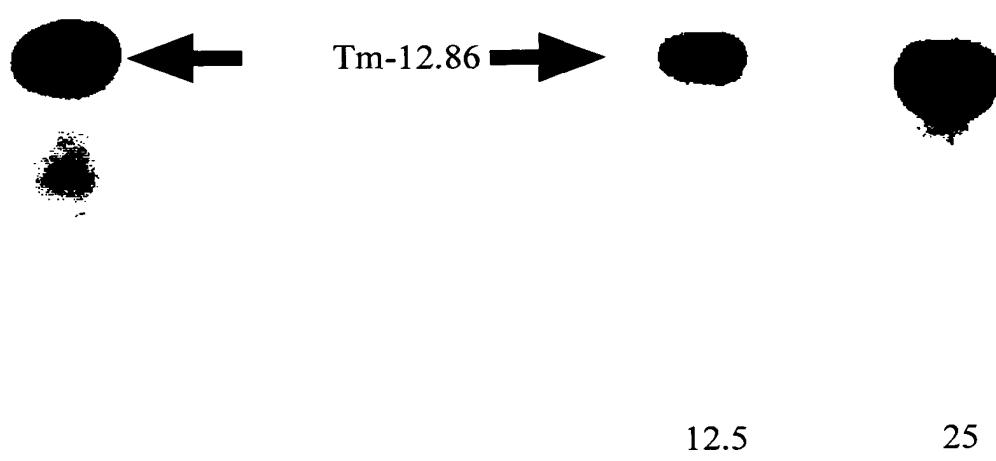


Fig. 1.3

Fig. 1.4

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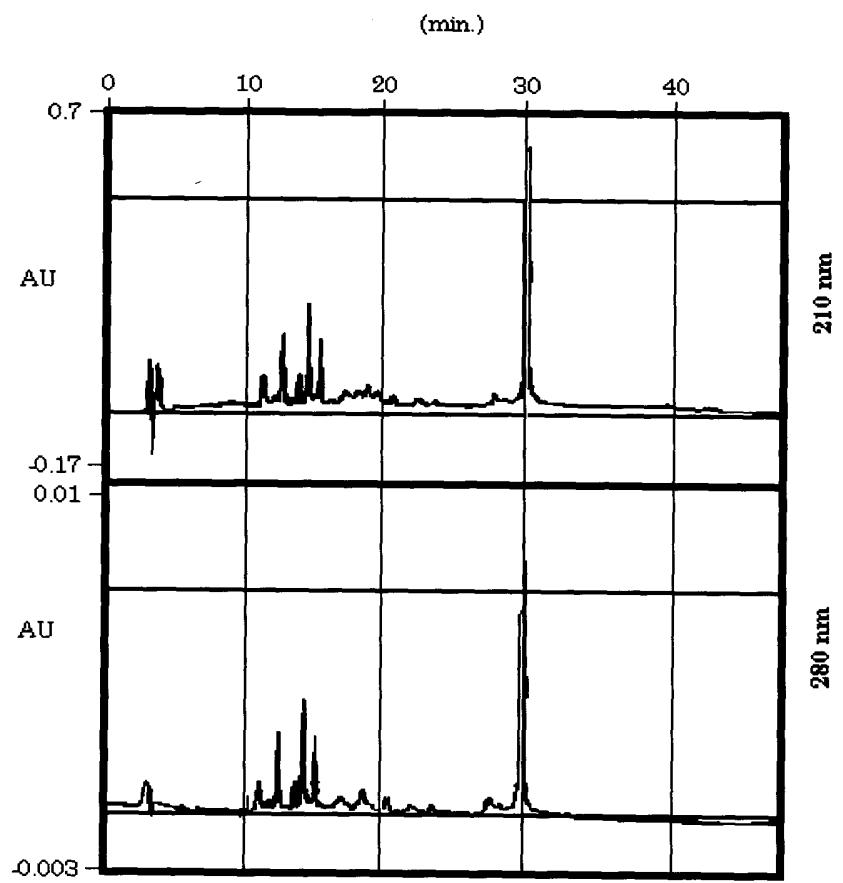


Fig. 1.5

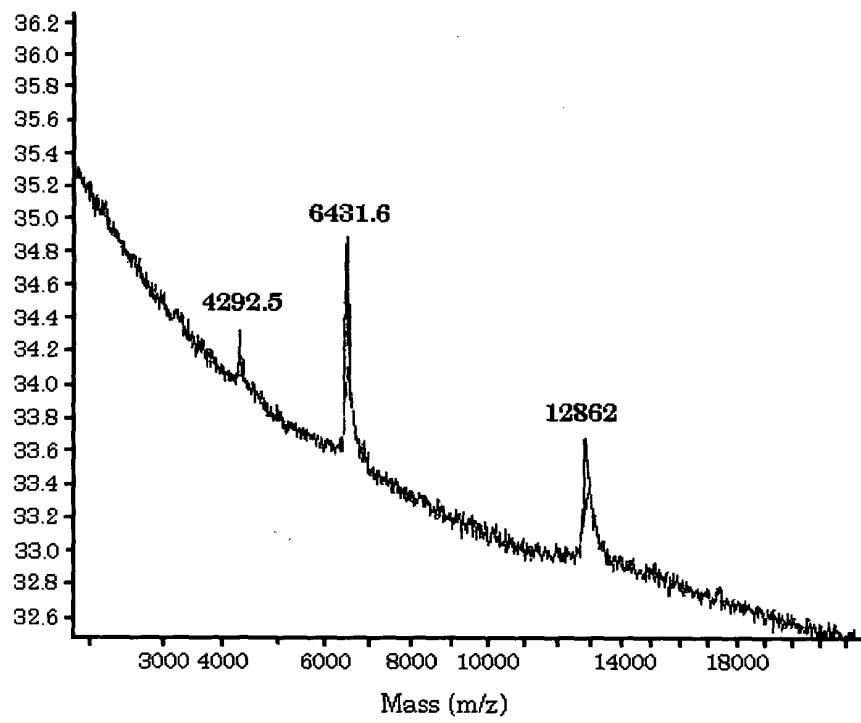


Fig. 1.6

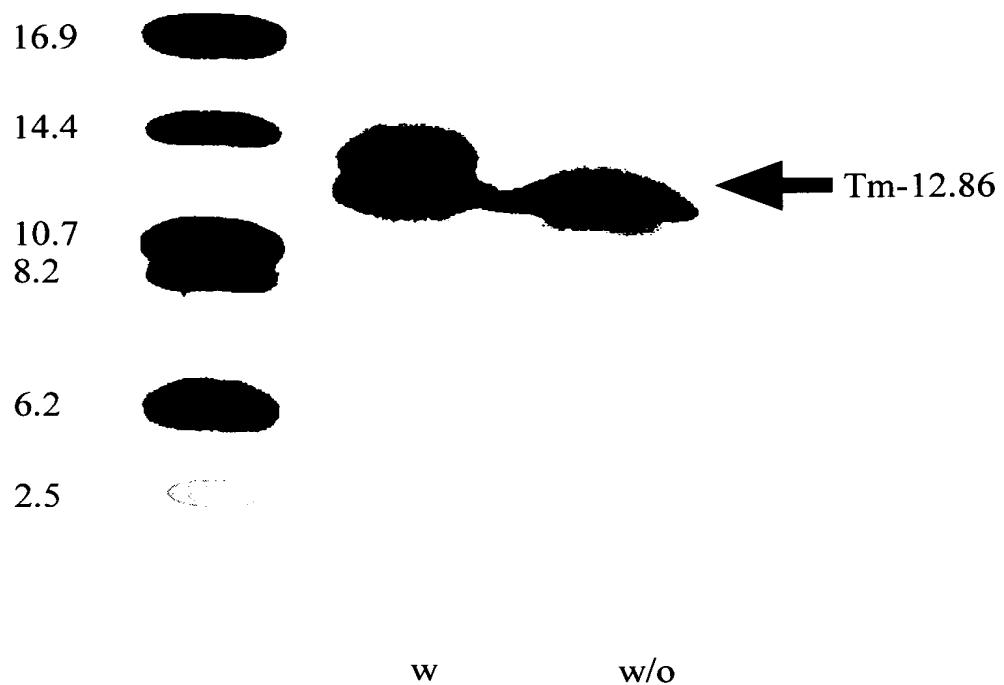


Fig. 1.7

NH₂-L-T-D-E-Q-I-Q-K-R-N-K-I-S-K-E-?-Q-Q-V

Val
Gln
Gln
Glu
Gln
Glu
Lys
Ser
Lys
Asn
Ile
Lys
Arg
Asn
Ile
Lys
Gln
Gln
Glu
Asp
Thr
Leu

Fig. 1.8

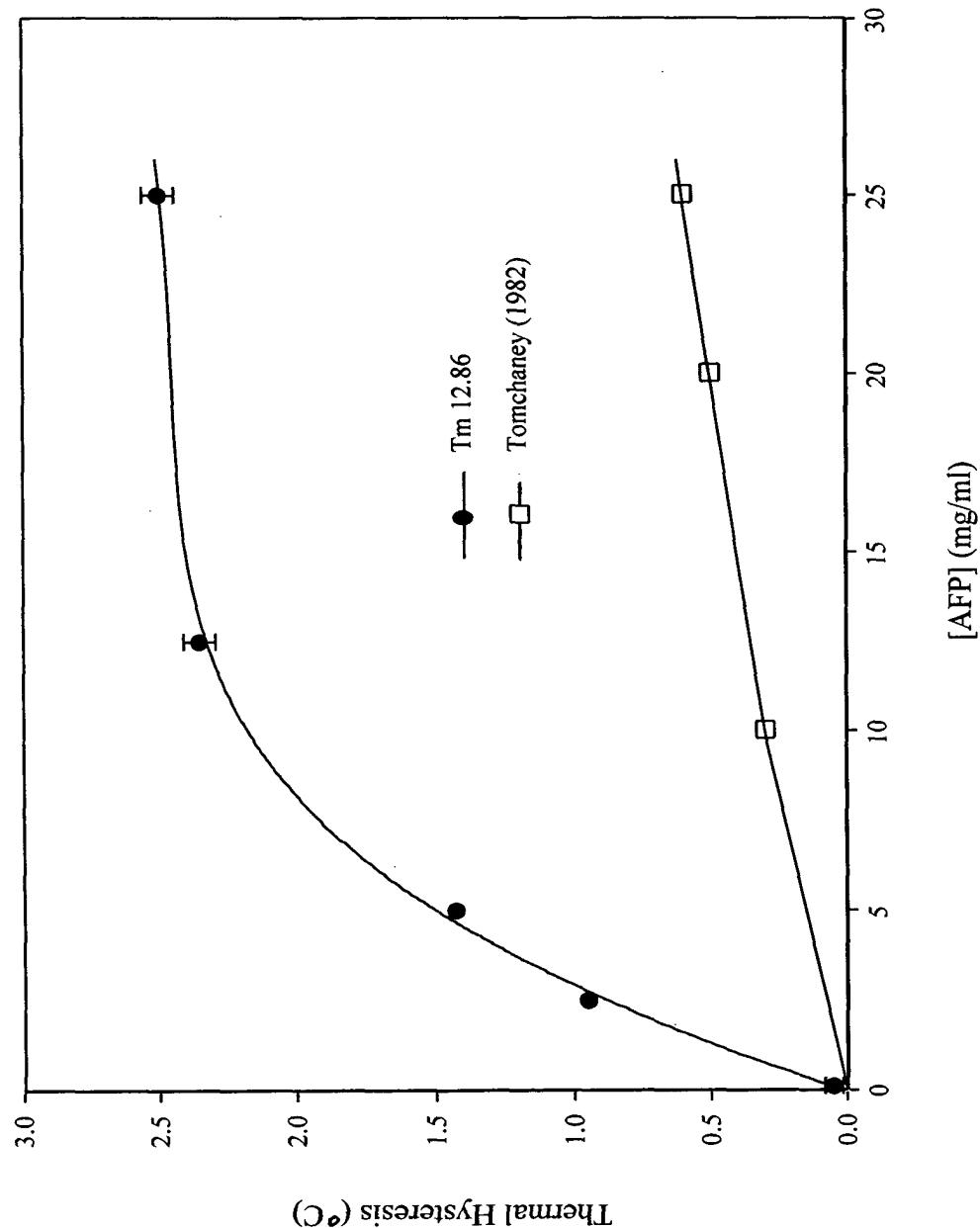


Fig. 1.9

NUCLEAR ENERGY

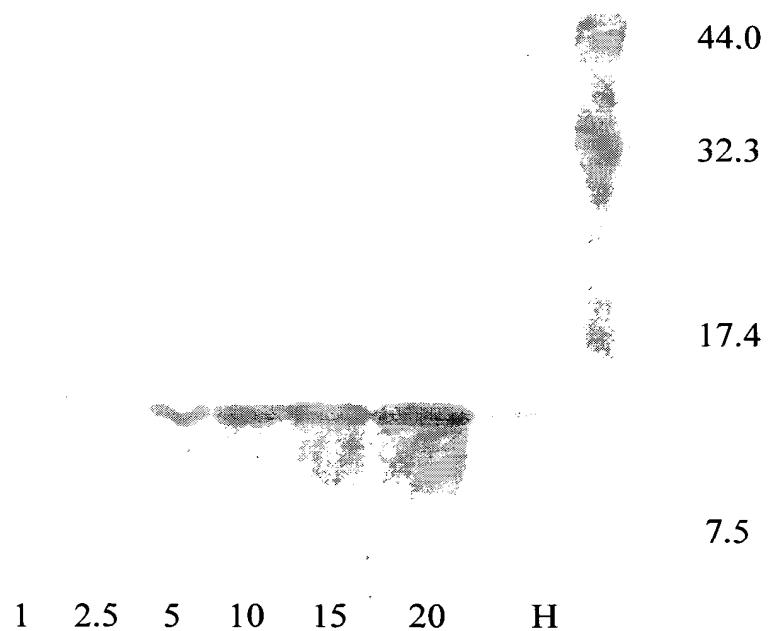


Fig. 1.10

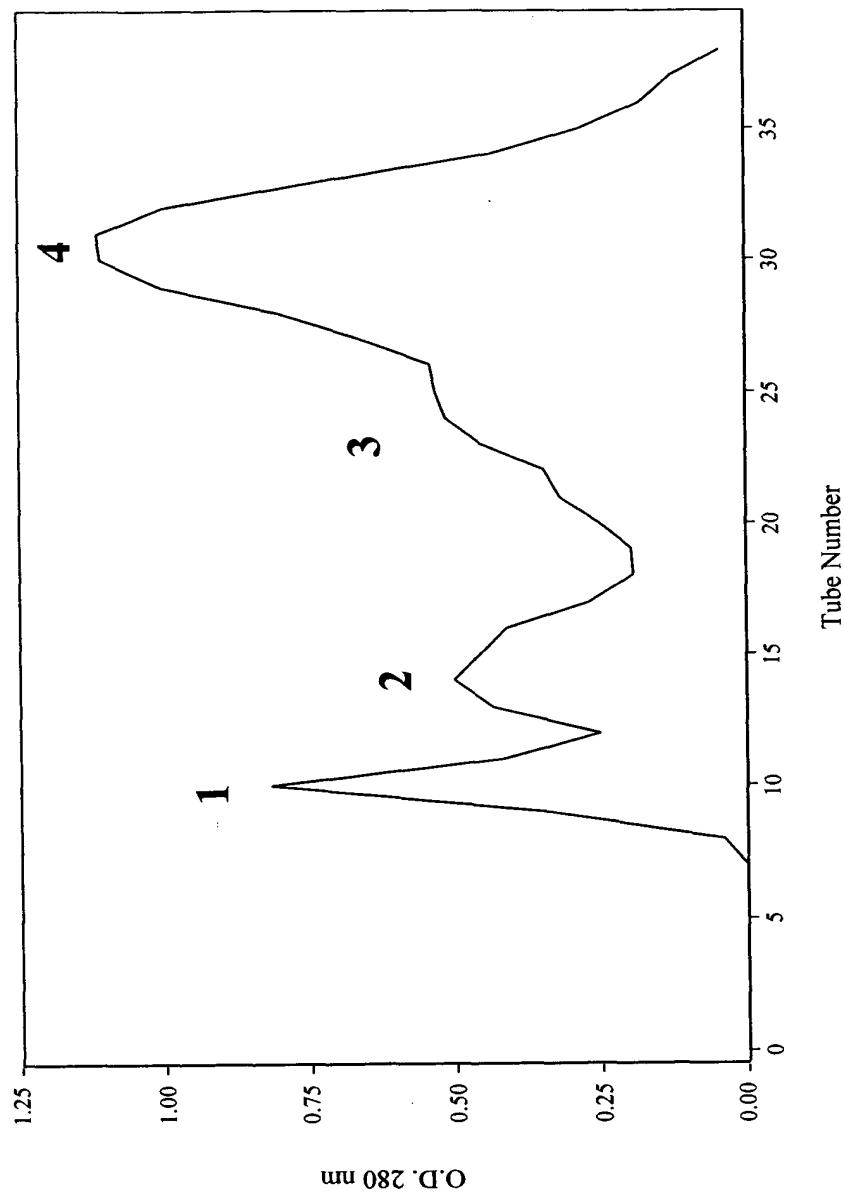


Fig. 1.11

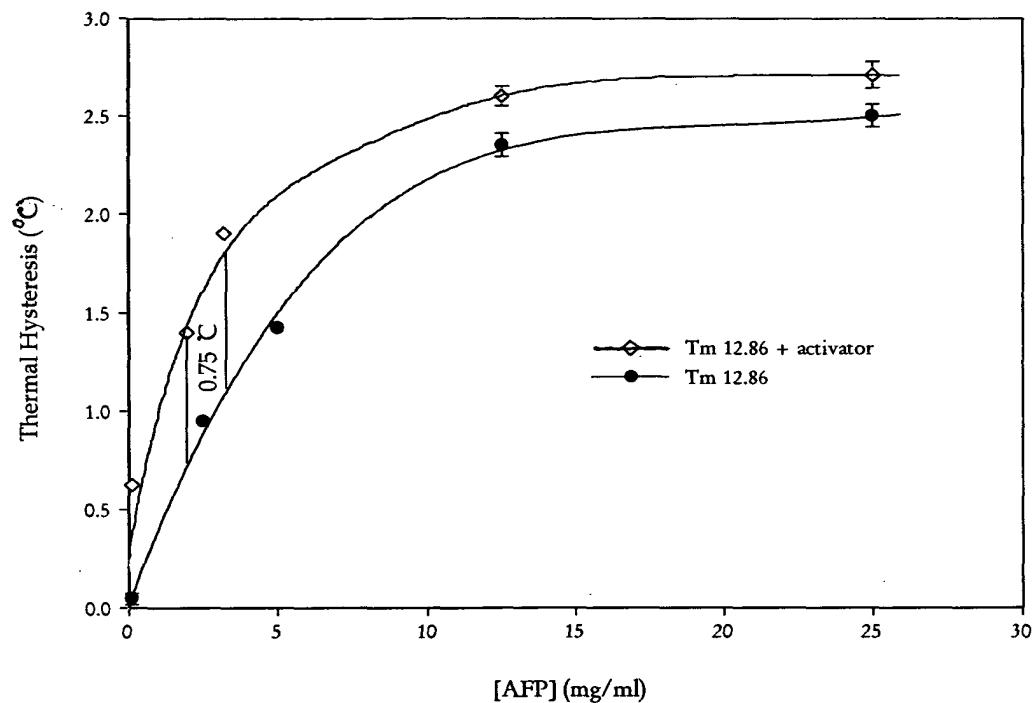


Fig. 1.12

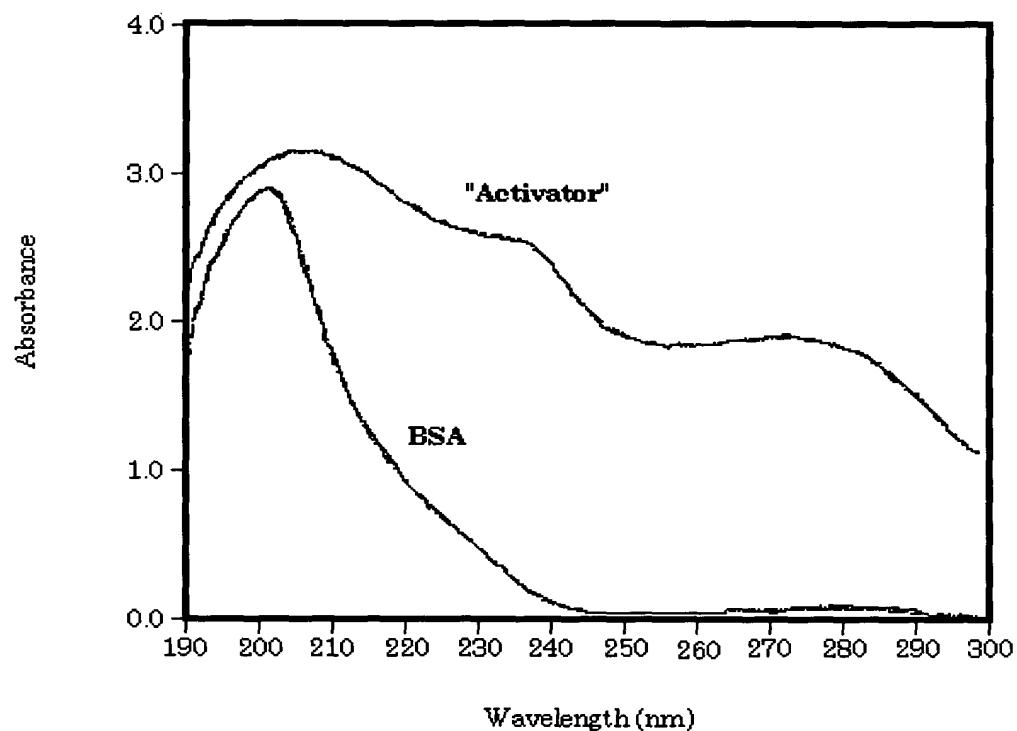


Fig. 1.13

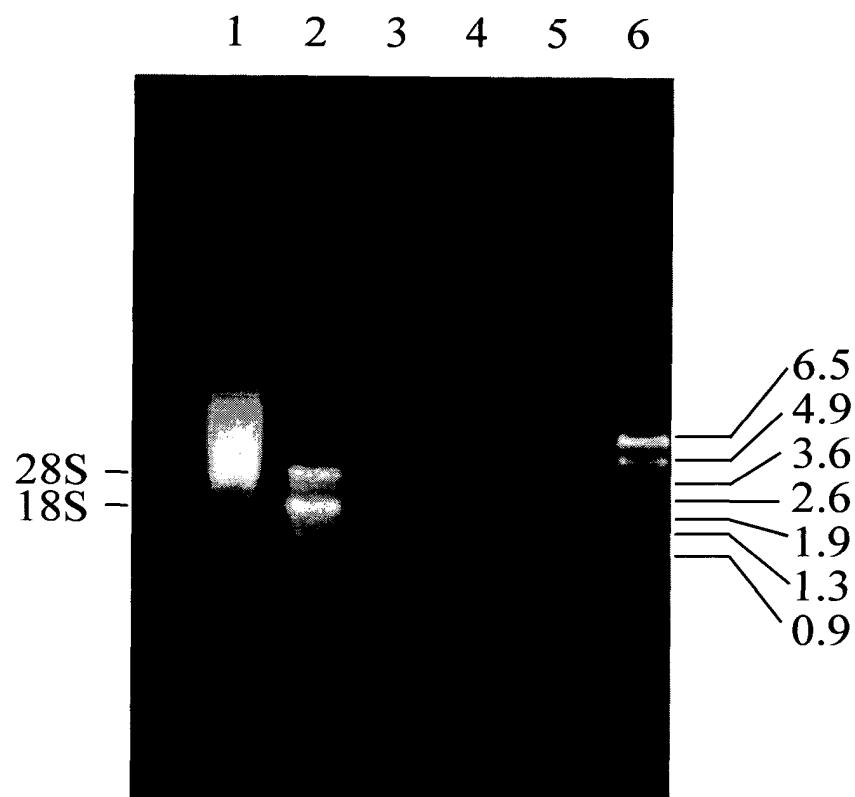


Fig. 2.0

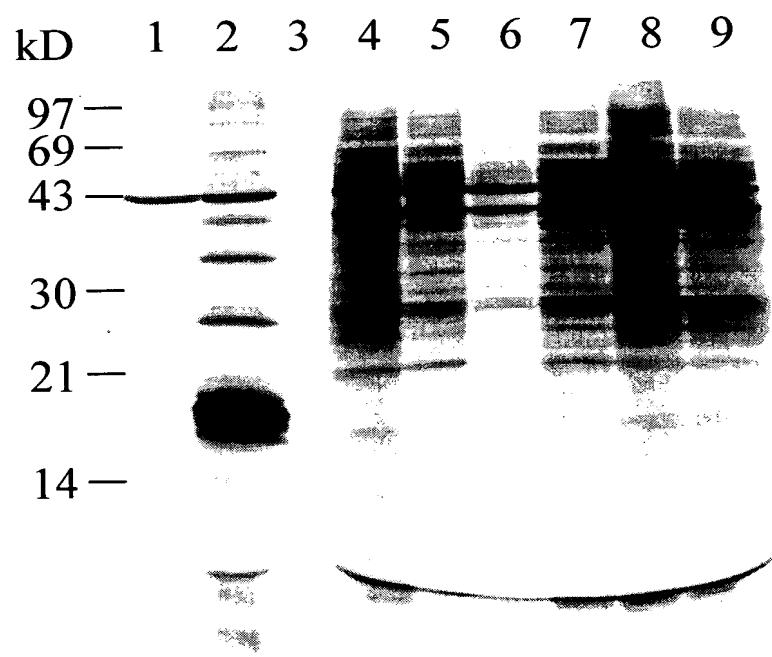


Fig. 2.1

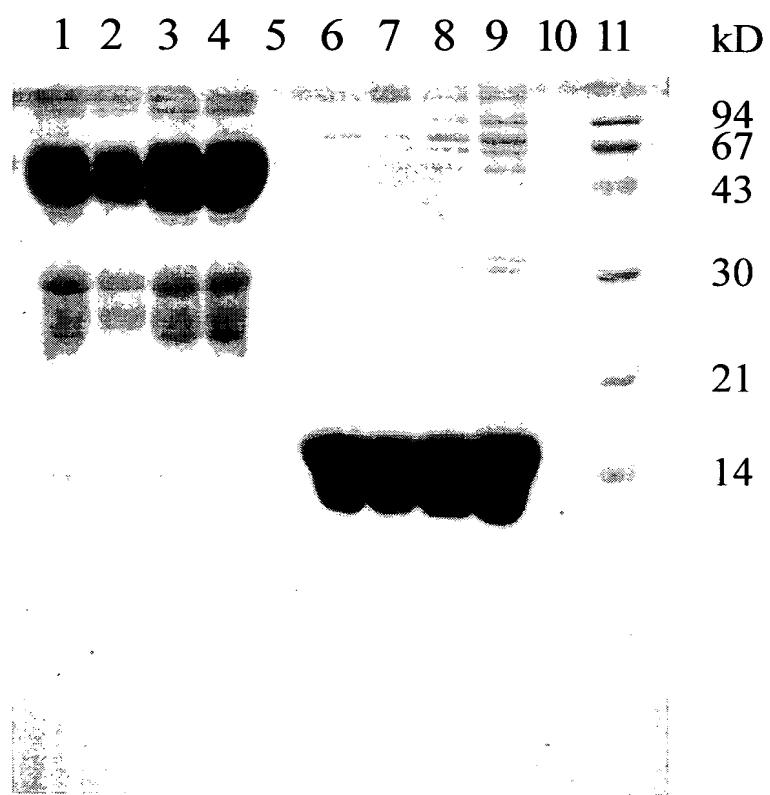


Fig. 2.2

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1 2 3 4 5 6 7 8 9 kD



Fig. 2.3

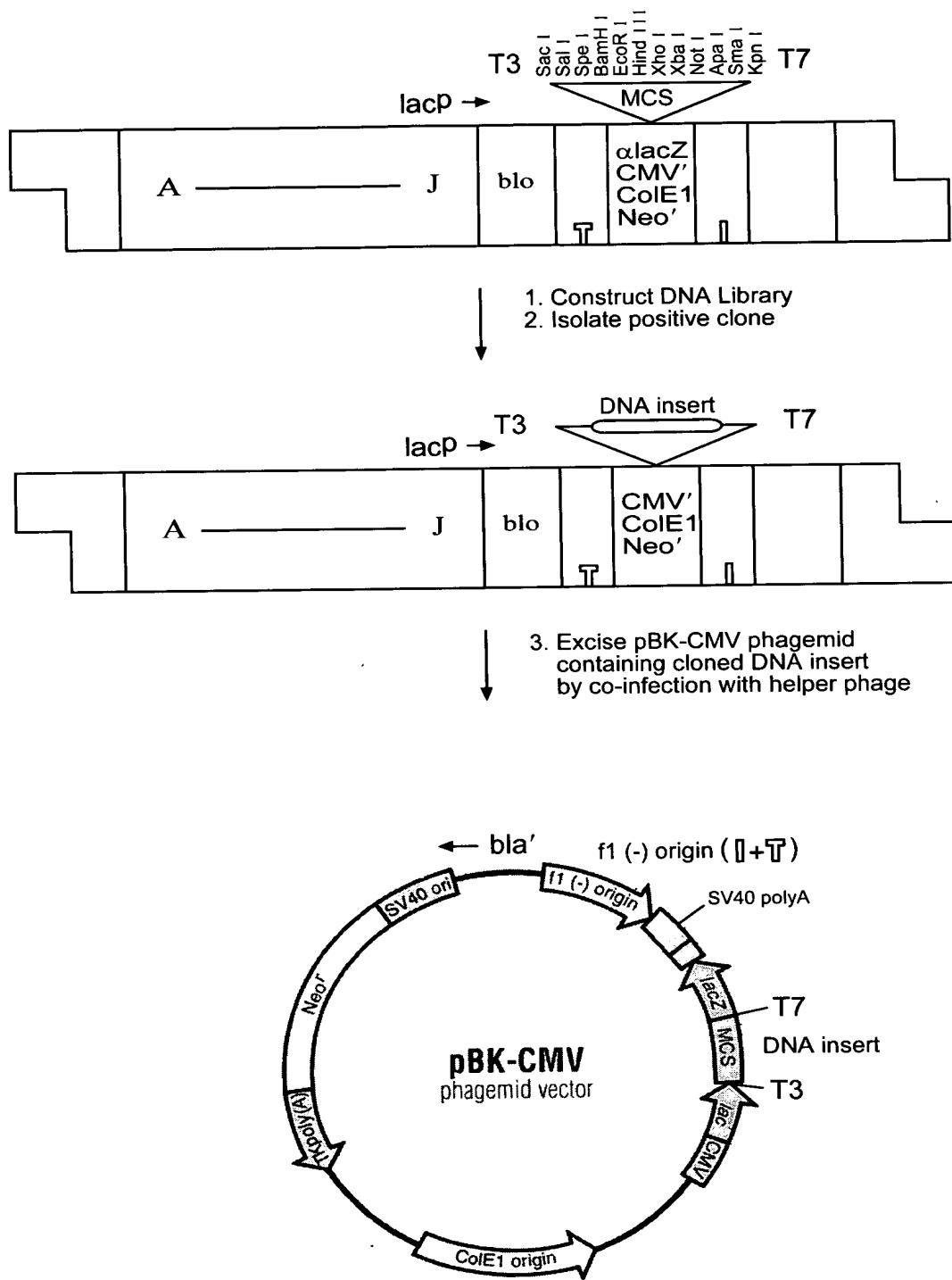
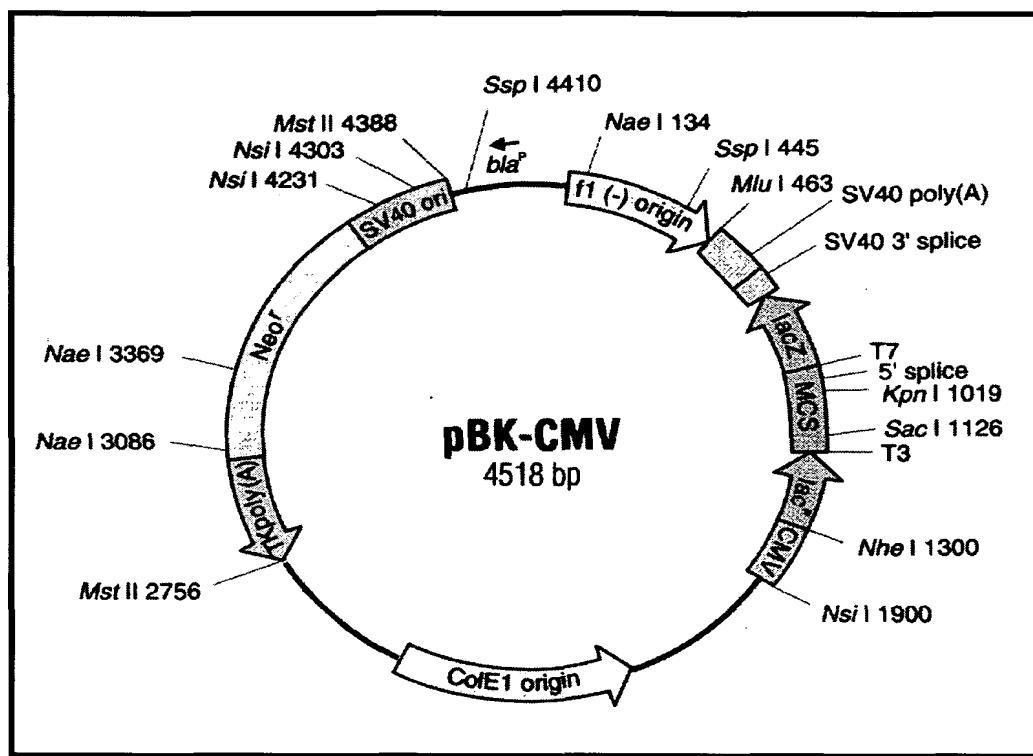


Fig. 2.4a



BK Reverse Primer
5' ACAGGAAACAGCTATGACCTTG 3'

1200 5' AATTAACCCCTCACTAAAGGG 3'
MET T3 promoter +1 →

1183 Sac I BssH II Sac I Sac I Bam H I Eco R I
T3 Reverse Primer
5' TCACACAGGAACAGCTATGACCTTGATTACGCCAACGTCGAAATTAAACCCCTCACTAAAGGGAACTAAAGGAACTGGAGCTGGCGCGCTGCAGGTGACACTAGTGGATCCAAG
3' AGTGTGTCCTTTGTCGATACTGGAAACTAATGGGGTTCGACCTTAATTGGGAGTGATTTCCTTTCGACCTCGAGCGCGGGAGCTGAGTCACCTAGTGGATCCAAG
↓
β-Galactosidase →

Hind III Sac I Xba I Not I Apa I Cla I Sac I Kpn I RspB06 I BstX I
ATTCAAAAAGCTCTCGAGAGTACTCTAGAGCGGCCGCGGCCCATCGATTTCCACCCGGGGGGTACCAAGGTAAAGTGTACCCAATTGCCCCATAGTGAAGTCGTTTACCAATTCACTGGCCGTGTTTACA 3' (+)
GTTTTGAAAGAGCTCTCATGAAGATECTCGCCGGGCCGGTAGCTAAAGGTGGGCCACCCCAAGGTGGGTTACATGGGTAAAGGGGATATCACTCAGCATAATGTTAAGTGACCCGGCGCAAAATGT 5' (-)
← +1 T7 promoter
3' CGGGATATCACTCAGCATAATG 5' TGACCCGGCGCAAAATG 5'
T7 Primer H13-20 Primer

Fig 2.46

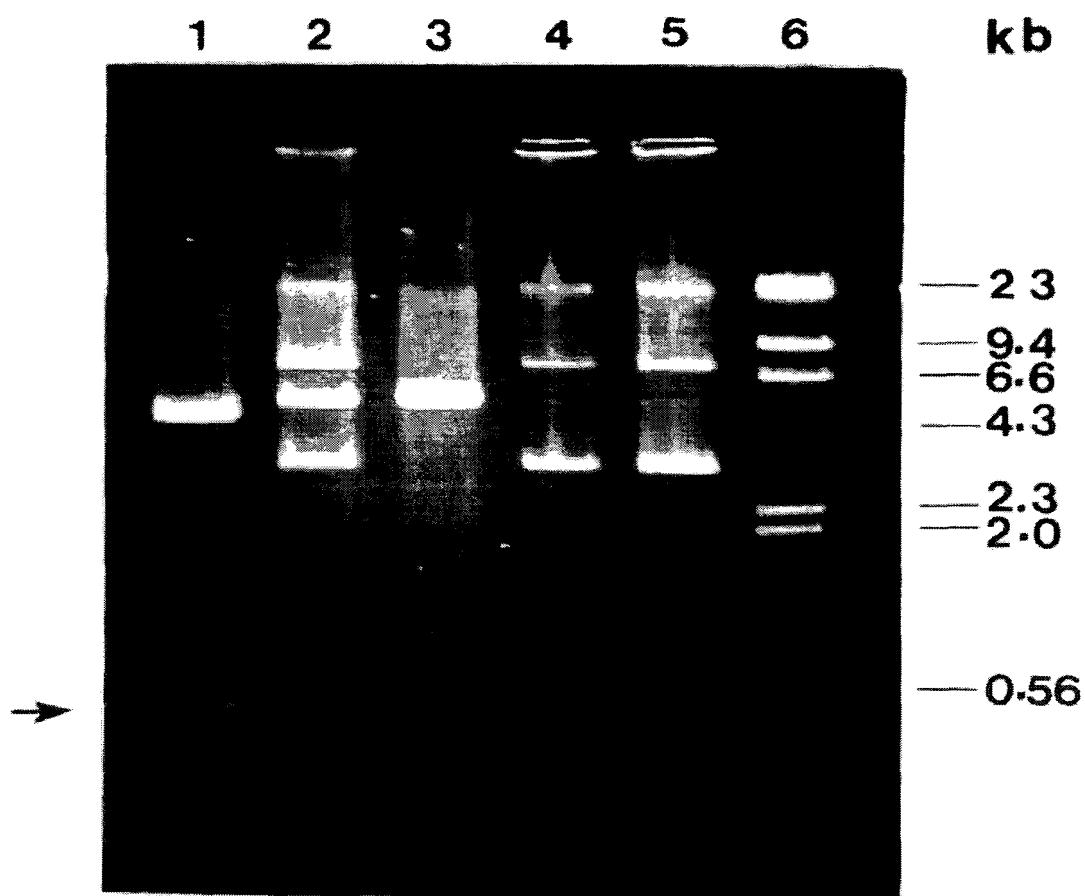


Fig. 2.5

DNA sequence of Tm 13.17 cDNA clone

B E
a C
m G
H R
I I

1 AGTGGATCCAAAGAATTGGCACGAGACTACTAAGTGAAGTTGCTCTGTTGTCTAATCT
M K L L C C L I S

61 CCCTCATTCCTGTTGGTCACAGTTCAGGCCCTGACCGAGGCACAAATTGAGAAACTGAACA
L I L L V T V Q A L T E A Q I E K L N K

121 AGATCAGCAAAAATGTCAAAATGAAAGTGGAGTGTCGAAGAGATCATAACCAAGCTC
I S K K C Q N E S G V S Q E I I T K A R

181 GCAACGGTGACTGGGAGGACGATCCTAAACTGAAACGCCAAGTTTTGGTGGCCAGGA
N G D W E D D P K L K R Q V F C V A R N

241 ACGCCGGTCTGGCCACGGAATCGGGAGAGGTGGTGGTCGACGTGTTGAGGGAGAACGTGA
A G L A T E S G E V V V D V L R E K V R

301 GGAAGGTCACTGACAACGACGAAGAAACTGAGAAAATCATCAATAAGTGCGCCGTCAAGA
K V T D N D E E T E K I I N K C A V K R

361 GAGATACTGTTGAAGAGAGACGGTGTCAATACTTCAAATGTGTCA
G A A A A A C A A G C C A A
D T V E E T V F N T F K C V M K N K P K

421 AGTTCTACCAGTTGATTGAACCACCACGACTAGTAGATGGTCAAATGGTGTGCTTAC
F S P V D *

X
h
o
i

481 ATATAAAAATAAAAGTGTCTGATGTAAAAAAAAAAAAAAAAAACTCG
polyadenylation signal poly (A) tail (26)

537 AGAGTATTCTAGAGCGGCCGGCCCATCGTTTCCACCC

Fig. 2.6a

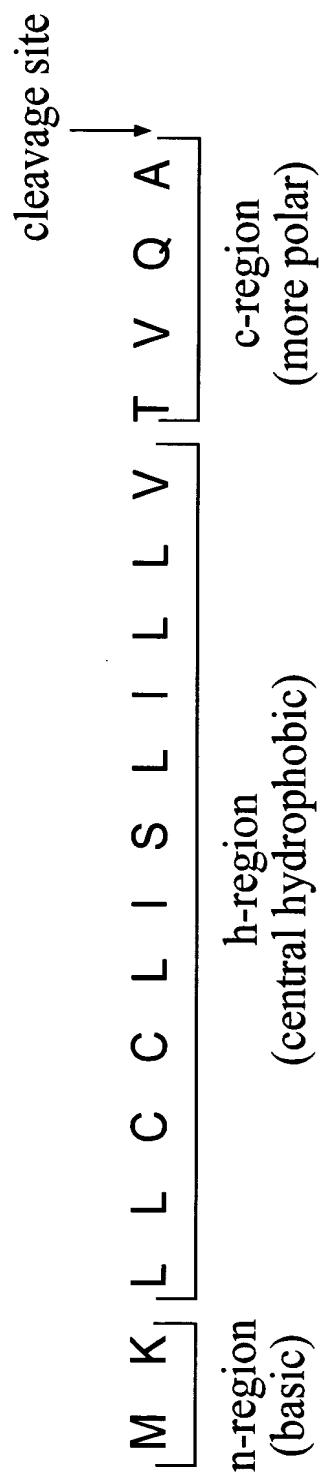


Fig. 2.6d

A. Mature Tm 13.17 amino acid residue

1 LTEAQIEKLN KISKKCQNES GVSQEIIITKA RNGDWEDDPK LKRQVFCVAR
51 NAGLATESGE VVVDVLREKV RKVTDNDEET EKIINKCAVK RDTVEETVFN
101 TFKCVMKNKP KFSPVD

B. Summary of the composition analysis for the mature Tm 13.17 sequence:

<u>Residue</u>	<u>Number</u>	<u>Mole Percent</u>
A = Ala	6	5.172
B = Asx	0	0.000
C = Cys	4	3.448
D = Asp	8	6.897
E = Glu	13	11.207
F = Phe	4	3.448
G = Gly	4	3.448
H = His	0	0.000
I = Ile	6	5.172
K = Lys	16	13.793
L = Leu	5	4.310
M = Met	1	0.862
N = Asn	8	6.897
P = Pro	3	2.586
Q = Gln	4	3.448
R = Arg	6	5.172
S = Ser	5	4.310
T = Thr	8	6.897
V = Val	14	12.069
W = Trp	1	0.862
Y = Tyr	0	0.000
Z = Glx	0	0.000

Molecular weight = 13171.96; Residues = 116; Average Residue Weight = 113.551

Charge = 1; Isoelectric point = 7.74.

Fig. 2.6c

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1	A T G G A T C C A R G A T T C G G C R C G R G A C T A G A T G A A	Tm 13.17 B 1
41	G T T O C T C T G T T D T C T A A T C T C C C T C T A T T C T G T T G G T C A C A C T T A C T T C T C T C T C A T T C T G T T G G T C C C A	Tm 13.17 B 1
81	G T T C A G G C C C T T A A C C O G G C A C A R A T T T G A O R A A T C T G A C A	Tm 13.17
28	G T T C A G G C C A T T A A C T C A D O R A A G G C C T T O A O C T A C T O C C C C	B 1
121	A G A T T C A G C A R A A A A T O T C A R A A A T T G A A G T G O R O T O T C G C A	Tm 13.17
68	A A A C C C A G C G C A G A O T O C A R A G A C T G A A O T G O R O T O T C G C A	B 1
161	A O O O A T C A T A A C C A A G C T C G C A R A C C O O T G A C T G G G G G O A C	Tm 13.17
118	A O O C O T C A T A A D A D O O C T C O C C A R A A G O T O A C T T G G G G O A C	B 1
201	G A T C C T T A A C T O R A A R C C O C C C A R O T T T T T G C O T O G C C C A O O A	Tm 13.17
148	G R C C C C C A A R A C T O R A A R A T O C A R A C T T C T T T G C A T T T T C R A O O	B 1
241	A C G C C C G O O T C T T O G C C C A C O G A T C G G G G A G G G T G G T O G T C G A	Tm 13.17
188	C A C T C C O A R A T T D T C G C C O A R T C G G G G A G G A T T O R O G C C C G A	B 1
281	C O T O T T D A D O O G A G A G O T G A D O A A G O O T C R A C T G A C A R A C O R C	Tm 13.17
228	C A C O T T C A R O O G A R A O T T O A C O A G O T O A C A R A C A G G A T G A T	B 1
321	G A A D A R A C T T O A C A R A A A T C A T C A R A T A G O T G C C C C O T C A R A O A	Tm 13.17
268	G A A D A R A A G C O A G A A O T T O T C O A G A A G O T O C A C G C O T G A C T O	B 1
361	G A A D A T A C T G T T G A R G A D O A C C G G T G T T C A R A T A C T T T C A R A T O	Tm 13.17
308	A A C C C C C A C T C C G G O A R A T A C G C C A T T T O A A D T T A C C A R A T O	B 1
401	T O T C A T O R A A A A R C A G C C C A R A O T T C T C T C A C C C A G T T G A T T O A	Tm 13.17
348	T O T A T T O R A B O G C A A O C C C A A T T T C T C A C C C A G T T G A T T O A	B 1
441	A C C A C C A C G C A T G A T G A T G T G T C A A A T G O O T G T G C T T T T A C	Tm 13.17
388	A G C A C C A A G C T A T T T G C T G R A A A A A A A A A A A A A A A A A A A A A A A A	B 1
481	A T A T A T A A R A T A A A O T G T T T C T G A T G T A R A A A A A A A A A A A A A A A A A A	Tm 13.17
428	G C A C C A A	B 1

Fig. 2.7

G E C E S E S - D A M B E R

Tm 13.17	3	EAQIEKLNKISKKCQNESGVSQEIITKARNGDWEDDPKLKRQVFCVARNA	52
	 : : .. : : ..	
AFP-3	1	ETPREKLKQHSDACKAESGVSEESLNKVRNREEVDDPKLKEHAFCILKRA	50
Tm 13.17	53	GLATESGEVVVDVLREKVRKVTDNDEETEKIINKCAVKRDTVEETVFNTF	102
		: .. . : : : . . . :	
AFP-3	51	GFIDASGEFQLDHIKTKFKENSEHPEKVDDLVAKCAVKKDTPQHSSADFF	100
Tm 13.17	103	KCVMKNKP	110
		. : .	
AFP-3	101	KCVHDNRS	108

Percent identity: 39.8 (identical amino acids); Percent similarity: 58.3 (identical amino acids plus conservative amino acids).

Fig. 2.8

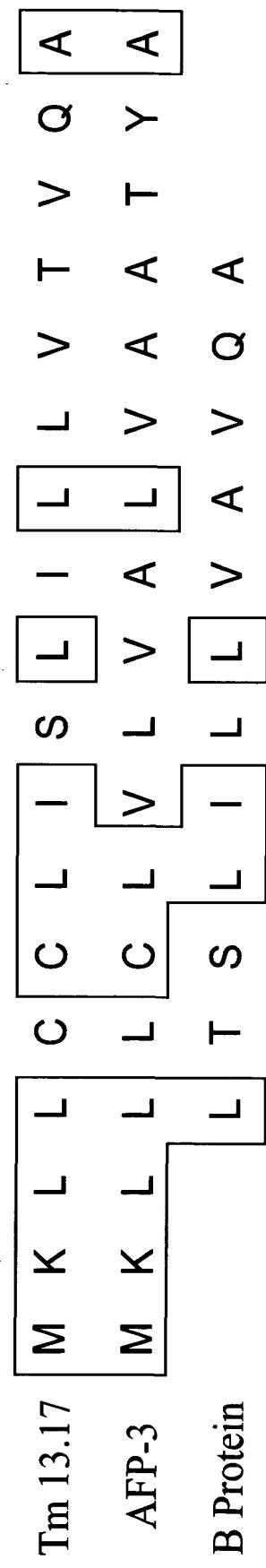


Fig. 2.9

TM 13.17	NH2-L T E A Q I E K L N K I S K K C Q N E
TM 12.86	NH2-L T D E Q I Q K R N K I S K E ? Q Q V

Fig 2.10

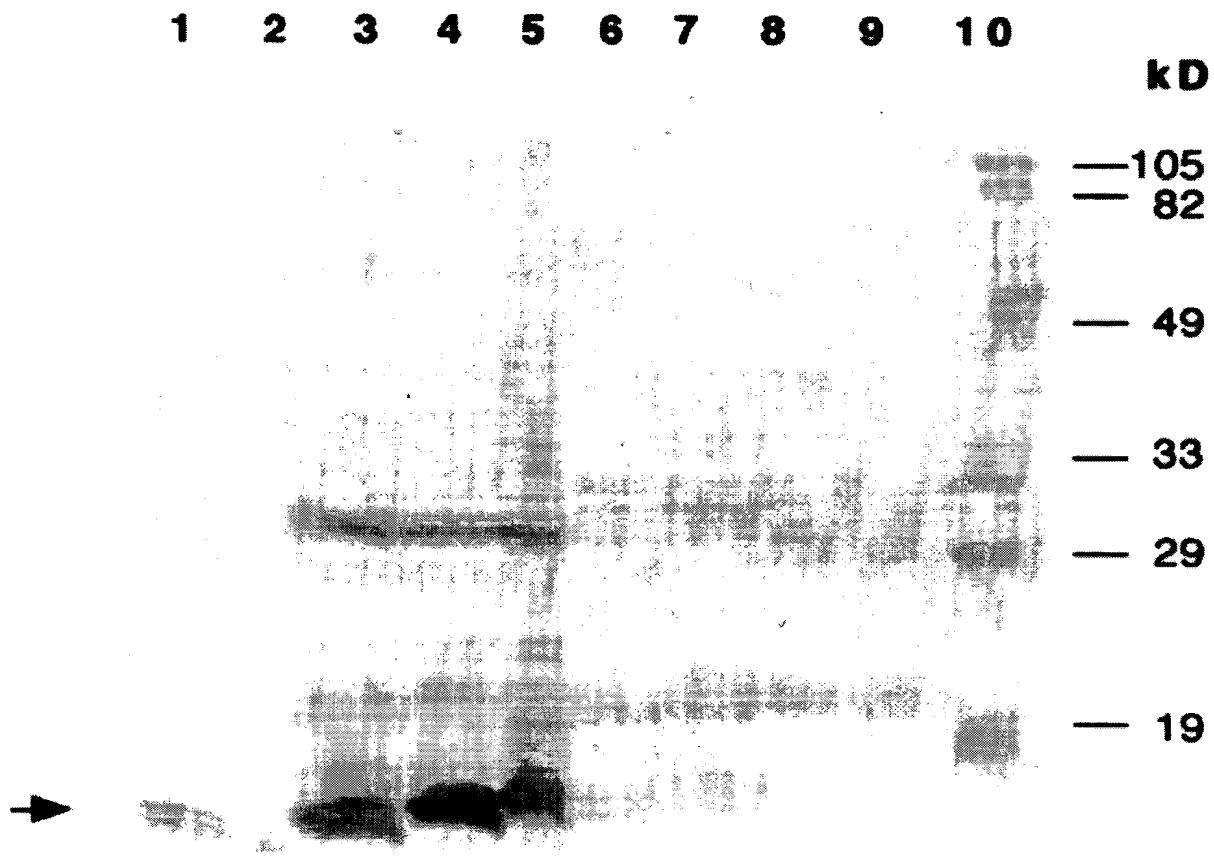


Fig 2.11

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Tm 12.86		L T D E Q I Q K R N K I S K E ? Q Q V
Tm 13.17	1	L T E A Q I E K L N K I S K K C Q N E S G V S Q E I I T K A
B1	13	I T E E D L E L L R Q T S A E C K T E S G V S E D V I K R A
AFP-3	1	E T P R E K L K Q H S D A C K A E S G V S E E S L N K V
Tm13.17	31	R N G D W E D D P K L K R Q V F C V A R N A G L A T E S G E
B1	44	R K G D L E D D P K L K M Q L L C I F K A L E I V A E S G E
AFP-3	29	R N R E E V D D P K L K E H A F C I L K R A G F I D A S G E
Tm13.17	61	V V V D V L R E K V R K V T D N D E E T E K I I N K C A V K
B1	75	I E A D T F K E K L T R V T N D D E E S E K I V E K C T V T
AFP-3	59	F Q L D H I K T K F K E N S E H P E K V D D L V A K C A V K
Tm13.17	91	R D T V E E T V F N T F K C V M K N K P K F S P V D
B1	106	E D T P E D T A F E V T K C V L K D K P N F F G D L F V
AFP-3	89	K D T P Q H S S A D F F K C V H D N R S

Fig. 2.12

1 GGCACGAGCAAAAATGAAACTCCCTTTGTGCTTGCGTCGCC
M K L L L C F A F A A

47 ATCGTCATCGGAGCTCAGGCTCTCACCGACGAACAGATACAGAAA
I V I G A Q A L T D E Q I Q K ↑

92 AGGAACAAGATCAGCAAAGAATGCCAGCAGGTGTCCGGAGTGTCC
R N K I S K E C Q Q V S G V S

137 CAAGAGACGATCGACAAAGTCCGCACAGGTGTCTGGTCATGAT
Q E T I D K Y R T G V L V D D

182 CCCAAAATGAAGAACGACGTCCCTCTGCTTCTCGAAGAAACTGGA
P K M K H Y L C F S K K T G

226 GTGGCAACCGAAGCCGGAGACACCAATGTGGAGGTACTCAAAGCC
V A T E A G D T N V E V L K A

271 AAGCTGAAGCATGTGGCCAGCGACGAAGAGGTGGACAAGATCGTG
K L K H V A S D E E V D K I V

316 CAGAAGTGCGTGGTCAAGAACGAGGCCACACCAAGAGGAAACGGCTTAT
Q K C V V K K A T P E E T A Y

361 GACACCTTCAAGTGTATTTACGACAGCAAAACCTGATTCTCTCCT
D T F K C I Y D S K P D F S P

406 ATTGATTAATTGTTTGATTTGACTGAATTTGACAATAAAAGGT
I D

polyadenylation signal

451 ACTATCGTTATGTAAAAAAAAAAAAAAA

poly (A) tail

Fig. 3.0

1 GGCACGAGCAAAAATGAAACTCCTCTTGTGCTTGGCTTCGCCGCC
 M K L L L C F A F A A

47 ATCGTCATCGGAGCTCAGGCTCTCACCGACGAACAGATAACAGAAA
 I V I G A Q A L T D E Q I Q K ↑

92 AGGAACAAGATCAGCAAAGAATGCCAGCAGGTGTCCGGAGTGTCC
 R N K I S K E C Q Q V S G V S

137 CAAGAGACGATCGACAAAGTCCGCACAGGTGTCTGGTCGACGAT
 Q E T I D K V R T G V L V D D

182 CCCAAAATGAAGAACGACGTCCCTCTGCTTCTCGAAGAAAAACTGGA
 P K M K K H V L C F S K K T G

226 GTGGCAACCGAACGCCGGAGACACCAATGTGGAGGTACTCAAAGCC
 V A T E A G D T N V E V L K A

271 AAGCTGAAGCATGTGGCCAGCGACGAAGAAAGTGGACAAGATCGTG
 K L K H V A S D E E V D K I V

316 CAGAAGTGCCTGGTCAAGAACGGCACACCCAGAGGAAACGGCTTAT
 Q K C V V K K A T P E E T A Y

361 GACACCTTCAAGTGTATTACGACAGTAAACCTGATTCTCTCCT
 D T F K C I Y D S K P D F S P

406 ATTGATTAATTGTTTGTATTTGACTGAATTTGACAAATAAGGT
 I D

polyadenylation signal

451 ACTATCGTTATGAAAAAAAAAAAAAA

poly (A) tail

Fig 3.1

Start

↓

2-2	GGCACGAGCAAAATGAAACTCCCTTGTGCTTTGC
2-3	GGCACGAGCAAAATGAAACTCCCTTGTGCTTTGCT
2-2	TTCGCCGCCATCGTCATCGGAGCTCAGGCTCTCACCG
2-3	TTCGCCGCCATCGTCATCGGAGCTCAGGCTCTCACCG
2-2	ACGAACAGATACAGAAAAGGAACAAGATCAGCAAAGA
2-3	ACGAACAGATACAGAAAAGGAACAAGATCAGCAAAGA
2-2	ATGCCAGCAGGTGTCCGGAGTGTCCCCAAGAGAGACGATC
2-3	ATGCCAGCAGGTGTCCGGAGTGTCCCCAAGAGAGACGATC
2-2	GACAAAGTCCGCACAGGTGTCTTGGTCGA[T]GATCCCCA
2-3	GACAAAGTCCGCACAGGTGTCTTGGTCGA[C]GATCCCCA
2-2	AAATGAAGAAAGCACGTCCCTCTGCTTCTCGAAGAAAAAC
2-3	AAATGAAGAAAGCACGTCCCTCTGCTTCTCGAAGAAAAAC
2-2	TGGAGTGGCAACCGAACGCCGGAGACACCAATGTGGAG
2-3	TGGAGTGGCAACCGAACGCCGGAGACACCAATGTGGAG
2-2	GTACTCAAAGCCAAGCTGAAGCATGTGGCCAGCGACG
2-3	GTACTCAAAGCCAAGCTGAAGCATGTGGCCAGCGACG
2-2	AAGAG[G]TGGACAAGATCGTGCAGAAGTGCCTGGTCAA
2-3	AAGA[A]GTGGACAAGATCGTGCAGAAGTGCCTGGTCAA
2-2	GAAGGCCACACCCAGAGGAAACGGCTTATGACACCTTC
2-3	GAAGGCCACACCCAGAGGAAACGGCTTATGACACCTTC
2-2	AAGTGTATTACGACAG[C]AAACCTGATTTCTCTCCTA
2-3	AAGTGTATTACGACAG[T]AAACCTGATTTCTCTCCTA
2-2	TTGATTAATTGTTTGATTTGACTGAATTTGACAA
2-3	TTGATTAATTGTTTGATTTGACTGAATTTGACAA
2-2	TAAAGGT[A]TATCGTTATG[T]AAAAAA
2-3	TAAAGGT[A]CTATCGTTATG[G]AAAAAA

Fig 3.2

Predicted Amino Acid Composition of 2-2 and 2-3

Analysis	Whole Protein
Molecular Weight	12843.80 m.w.
Length	115
1 microgram =	77.859 pMoles
Molar Extinction coefficient	3040±5%
1 A(280) =	4.22 mg/ml
Isoelectric Point	7.11
Charge at pH 7	0.13

Whole Protein Composition Analysis

Amino Acid(s)	Number count	% by weight	% by frequency
Charged (RKHYCDE)	48	47.19	41.74
Acidic (DE)	20	18.90	17.39
Basic (KR)	20	20.40	17.39
Polar (NCQSTY)	30	25.35	26.09
Hydrophobic (AILFWV)	34	27.26	29.57
A Ala	6	3.32	5.22
C Cys	4	3.21	3.48
D Asp	11	9.86	9.57
E Glu	9	9.05	7.83
F Phe	3	3.44	2.61
G Gly	4	1.78	3.48
H His	2	2.14	1.74
I Ile	6	5.29	5.22
K Lys	18	17.97	15.65
L Leu	5	4.41	4.35
M Met	1	1.02	0.87
N Asn	2	1.78	1.74
P Pro	4	3.02	3.48
Q Gln	6	5.98	5.22
R Arg	2	2.43	1.74
S Ser	7	4.75	6.09
T Thr	9	7.08	7.83
V Val	14	10.80	12.17
W Trp	0	0.00	0.00
Y Tyr	2	2.54	1.74
B Asx	0	0.00	0.00
Z Glx	0	0.00	0.00
X Xxx	0	0.00	0.00
Ter	0	0.00	0.00

Fig. 3.3

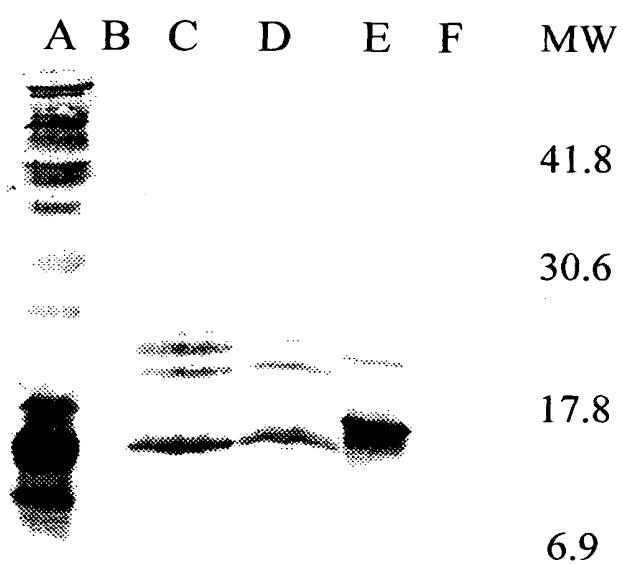


Fig. 3.4

2 3 4 5 6 7 8 9 10

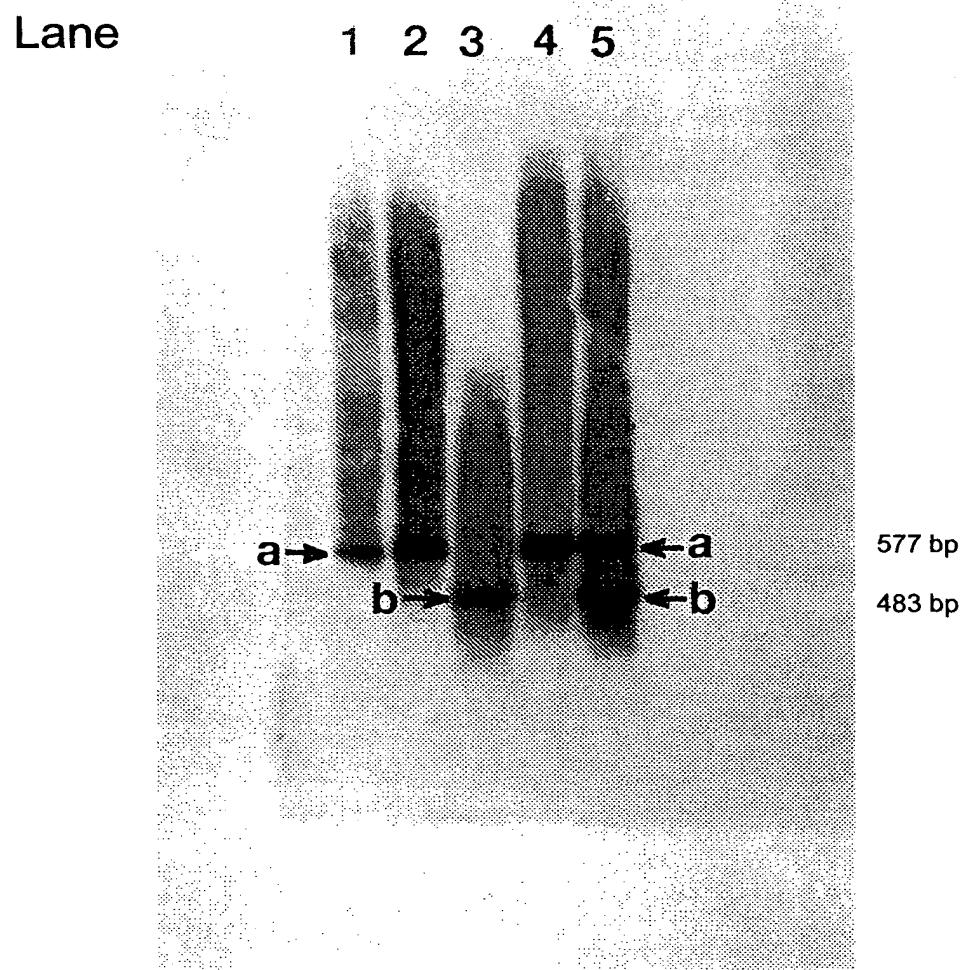


Fig. 4.0

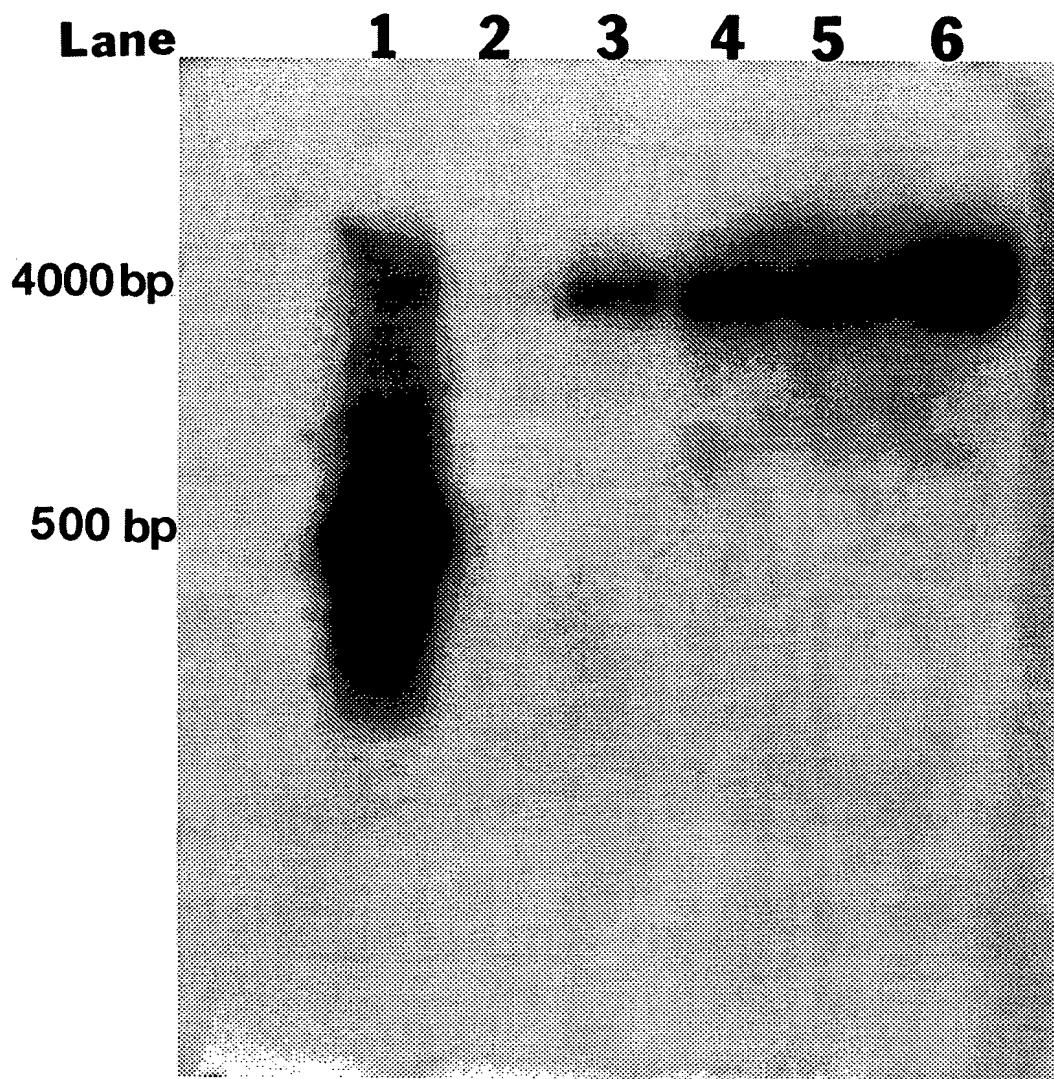


Fig. 4.1

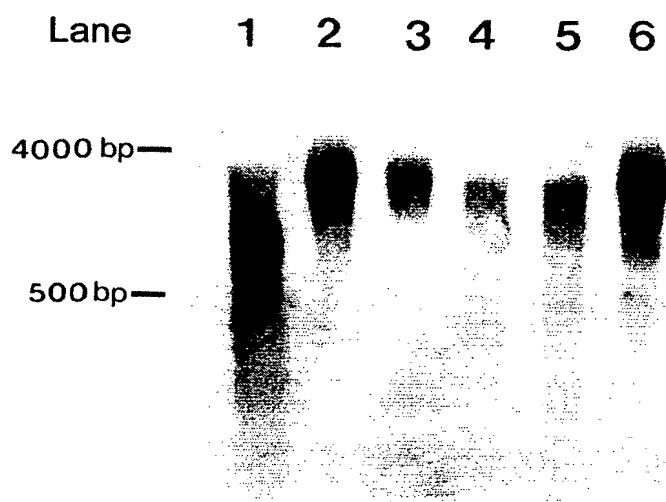


Fig. 4.2

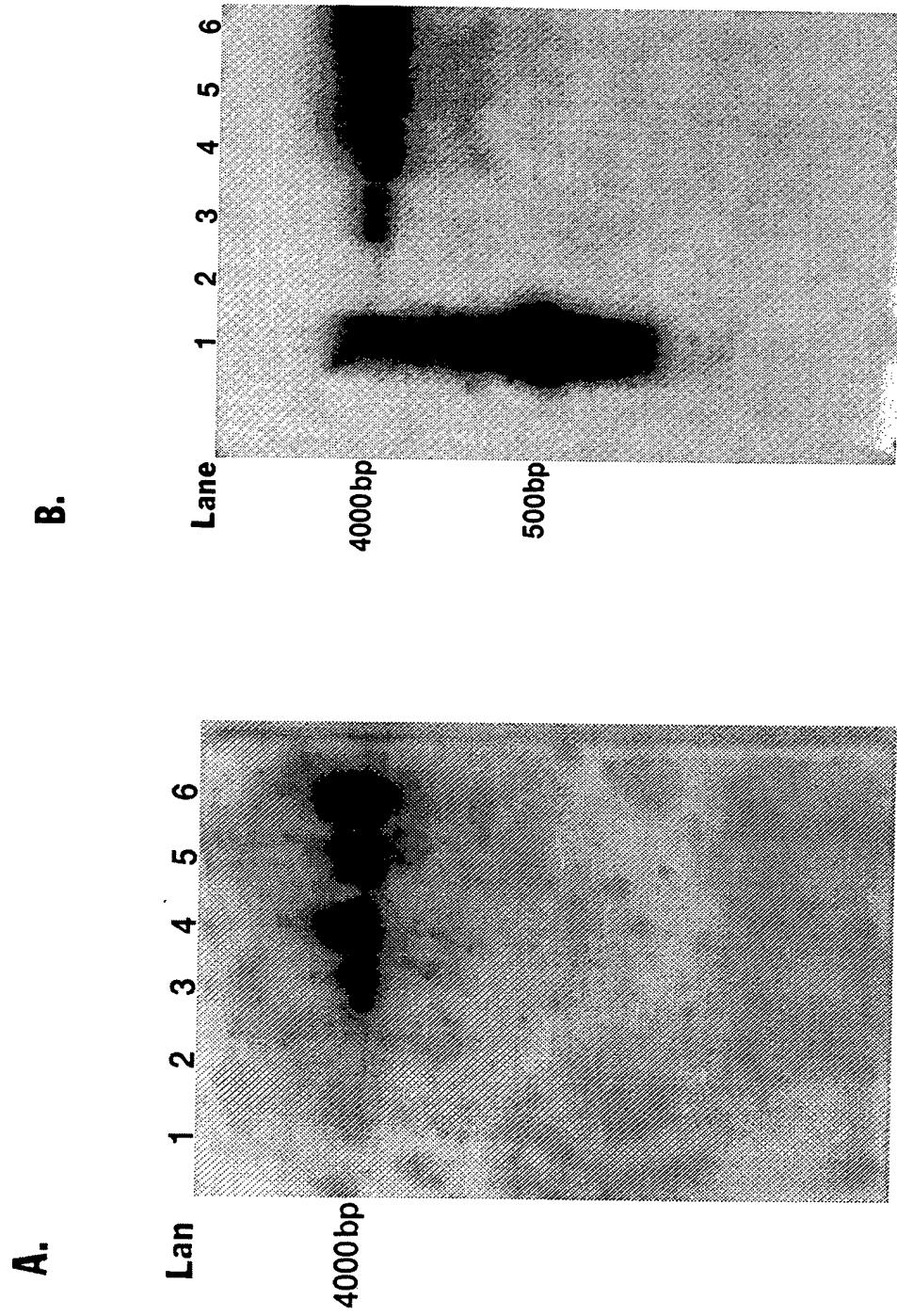


Fig. 4.3

— 4000 bp —

Lane 1 2 3 4 5 6

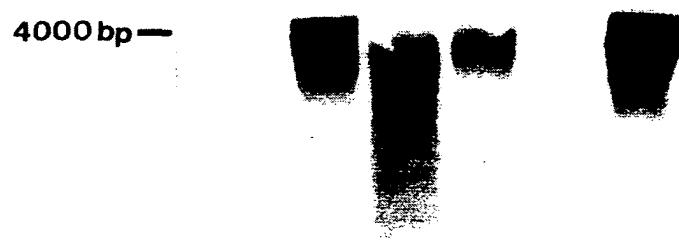


Fig. 4.4

Lane 1 2 3 4 5

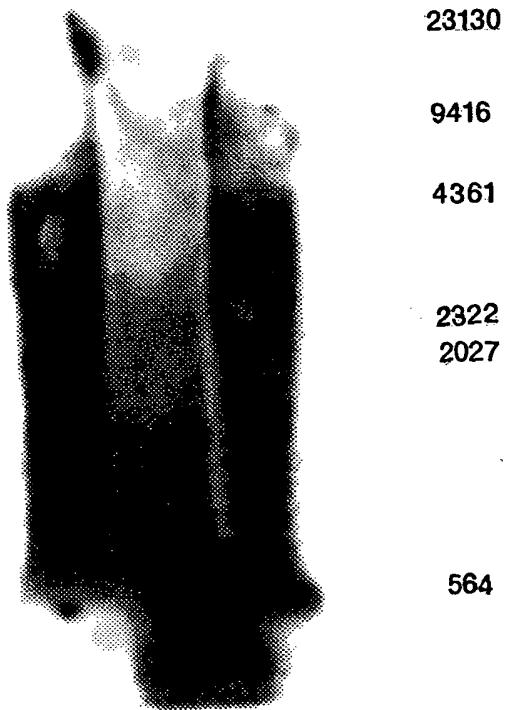


Fig. 4.5

Tm 13.17 cDNA

1 AGTGGATCCAAAGAATTGGCACGAGACTACTAAGAGATGAAGTTGCTCTGTTGTCTAATCT
M K L L C C L I S

61 CCCTCATTCTGTTGGTCACAGTTCAAGGCCCTGACCGAGGCACAAATTGAGAAAACTGAACA
L I L L V T V Q A L T E A Q I E K L N K
Forward Primer

121 AGATCAGCAAAAATGTCAAAATGAAAGTGGAGTGTCGCAAGAGATCATAACCAAGCTC
I S K K C Q N E S G V S Q E I I T K A R

181 GCAACGGTGACGGAGGACGATCCTAAACTGAAACGCCAAGTTTTGCGTGGCCAGGA
N G D W E D D P K L K R Q V F C V A R N

241 ACGCCGGTCTGGCCACGGAATGGGAGAGGTGGTGGTCGACGTGTTGAGGGAGAAGGTGA
A G L A T E S G E V V V D V L R E K V R

301 GGAAGGTCACTGACAACGACGAAGAAAACTGAGAAAATCATCAATAAGTGCGCCGTCAAGA
K V T D N D E E T E K I I N K C A V K R
Reverse Primer

361 GAGATATGTGAAGAGACGGTGTCAATATTTCAAATGTGTCATGAAAAACAAGCCAA
D T V E E T V F N T F K C V M K N K P K

421 AGTTCTCACCAGTTGATTGAACCACCACGACTAGTAGATGGTTCAAATGGTGTGCTTTAC
F S P V D *

481 ATATAAAAATAAAGTGTTCTGATGTAAAAAAAAAAAAAAAAACTCG
polyadenylation signal poly (A) tail (26)

537 AGAGTATTCTAGAGCGGCCGGGGCCCCATCGTTCCACCC

Fig. 4.6a

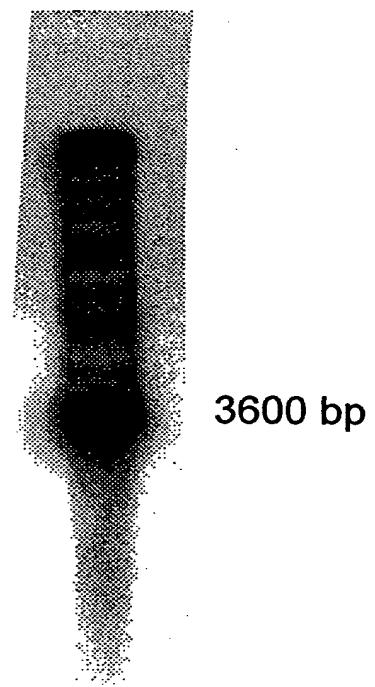
B2 Tm 13.17 B2 AFP-3

	Forward Primer
2-2	L T D E Q I Q K R N K I S K E C Q Q V S G V S Q E T I D K V R T G V L V
Tm 13.17	L T E A Q I E K L N K I S K K C Q N E S G V S Q E I I T K A R N G D W E
B2	L T E E D L Q L L R Q T S A E C K T E S G A S E A V I K K A R K G D L E
AFP-3	E T P R E K L K Q H S D A C K A E S G V S E E S L N K V R N R E E V
2-2	D D P K M K K H V L C F S K K T G V A T E A G D T N V E V L K A K L K H
Tm 13.17	D D P K L K R Q V F C V A R N A G L A T E S G E V V V D V L R E K V R K
B2	D D P K L K M Q L L C I F K A L E I V A E S G E I E A D T F K E K L T R
AFP-3	D D P K L K E H A F C I L K R A G F I D A S G E F Q L D H I K T K F K E
	Reverse Primer
2-2	V A S D E E V D K I V Q K C V V K K A T P E E T A Y D T F K C I Y D S
Tm 13.17	V T D N D E E T E K I I I N K C A V K R D T V E E T V F N T F K C V M K N
B2	V T N D D E E S E K I V E K C T V T E D T P E D T A F E V T K C V L K D
AFP-3	N S E H P E K V D D L V A K C A V K K D T P Q H S S A D F F K C V H D N

Fig. 4.6b

Primer	percent % composition				Melting Temperature (°C)
	A	C	G	T	
Forward	28.6	14.3	42.9	14.3	44.0
Reverse	25.0	31.3	6.3	37.5	44.0

Fig. 4.6c



3600 bp

Fig. 4.7

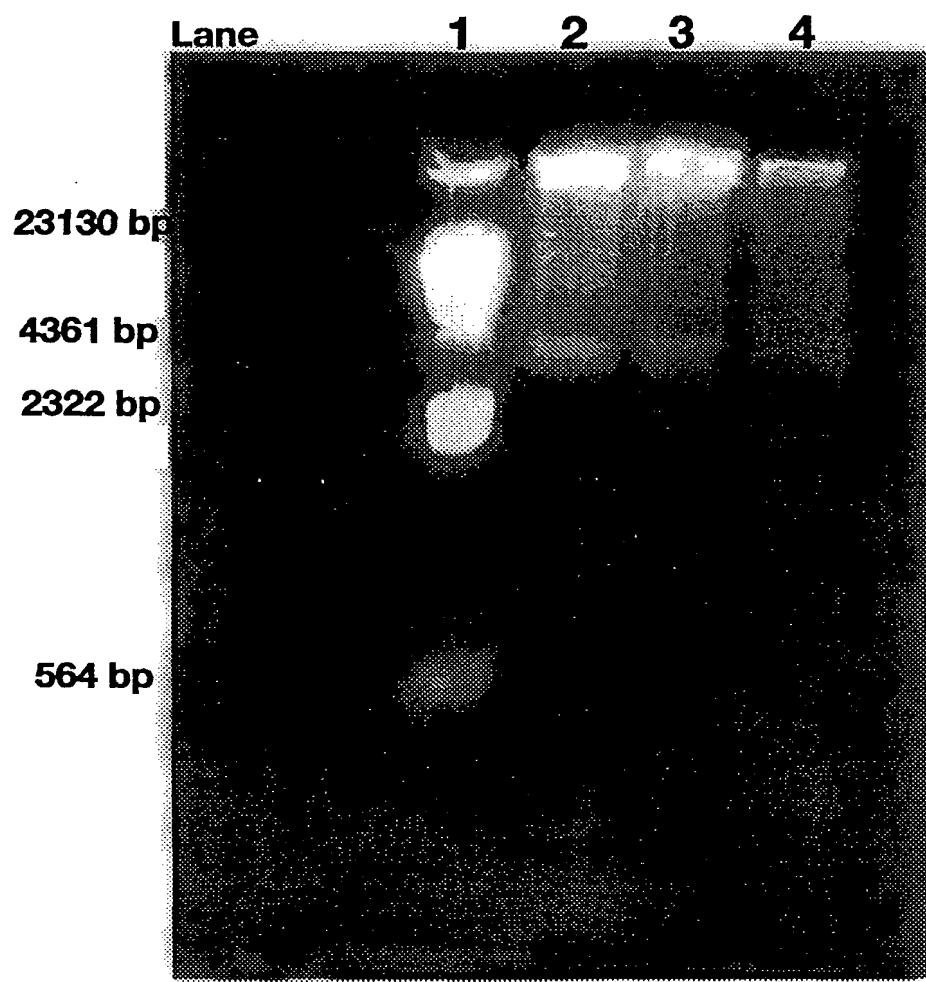


Fig. 4.8

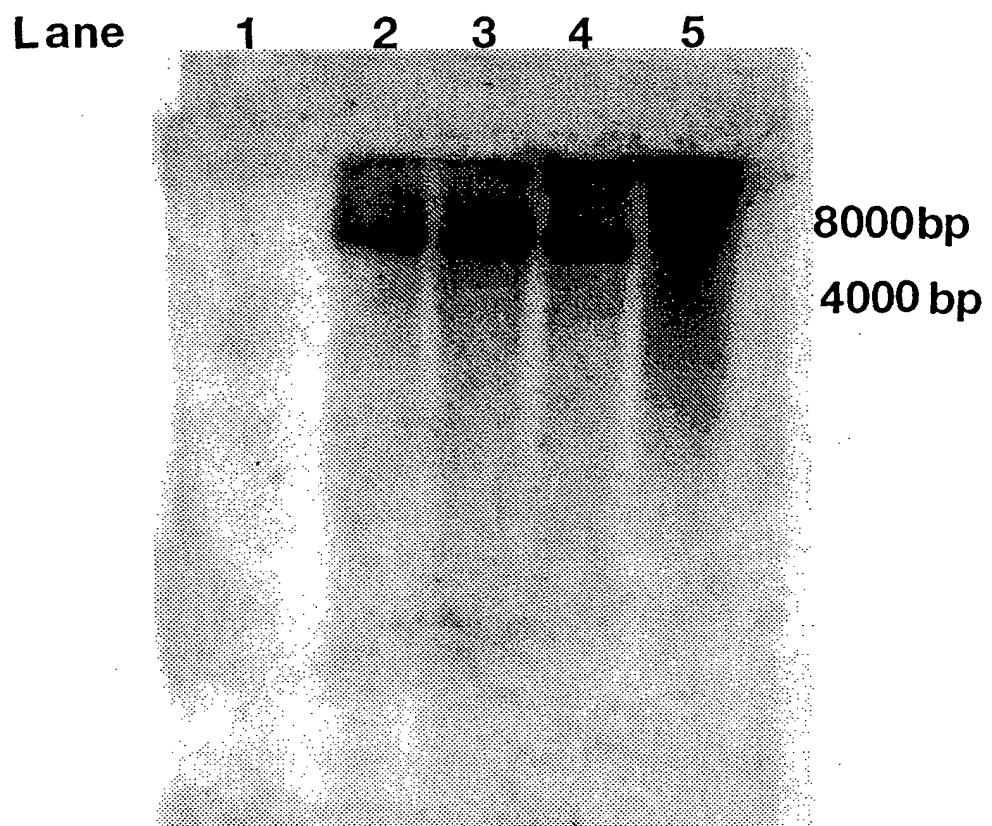


Fig. 4.9

47 ATCGTCATCGGAGCTCAGGCTCTCACCGACGAACAGATAACAGAAA
 I V I G A Q A L T D E Q I Q K

92 AGGAACAAAGATCAGCAAAGAATGCCAGCAGGGTGTCCGGAGTGTCC
 R N K I S K E C Q Q V S G V S

137 CAAGAGACGATCGACAAAGTCCGCACAGGTGTCTTGGTCGACGAT
 Q E T I D K V R T G V L V D D

182 CCCAAAAATGAAGAACGACGTCCCTCTGCTTCTCGAAGAAAATGGAA
 P K M K K H V L C F S K K T G

226 GTGGCAACCGAACGCCGGAGACACCAATGTGGAGGTACTCAAAGCC
 V A T E A G D T N V E V L K A

271 AAGCTGAAGCATGTGGCCAGCGACGAAGAGGGTGGACAAGATCGTG
 K L K H V A S D E E V D K I V

316 CAGAACGTGCGTGGTCAAGAACGCCACACCAAGAGGAAACGGCTTAT
 Q K C V V K K A T P E E T A Y

361 GACACCTCAAGGTTATTTACGACAGTAACCTGATTTCTCTCCT
 D T F K V I Y D S K P D F S P

406 ATTGATTAAATTGTTTGTATTTGACTGAATTGACAATAAAAGGT
 I D *

451 ACTATCGTTATGTAAAAAAAAAAAAAAAAAA

polyadenylation signal

Fig. 4.10a

Predicted Amino Acid

Composition of 3-4

Molecular Weight	12839.70 m.w.
Length	115
1 microgram =	77.883 pMoles
Molar Extinction coefficient	2920±5%
1 A(280) =	4.40 mg/ml
Isoelectric Point	7.14
Charge at pH 7	0.16

Whole Protein Composition Analysis

Amino Acid(s)	Number count	% by weight	% by frequency
Charged (RKHYCDE)	47	46.41	40.87
Acidic (DE)	20	18.91	17.39
Basic (KR)	20	20.41	17.39
Polar (NCQSTY)	29	24.55	25.22
Hydrophobic (AILFWV)	35	28.04	30.43
A Ala	6	3.32	5.22
C Cys	3	2.41	2.61
D Asp	11	9.86	9.57
E Glu	9	9.05	7.83
F Phe	3	3.44	2.61
G Gly	4	1.78	3.48
H His	2	2.14	1.74
I Ile	6	5.29	5.22
K Lys	18	17.97	15.65
L Leu	5	4.41	4.35
M Met	1	1.02	0.87
N Asn	2	1.78	1.74
P Pro	4	3.02	3.48
Q Gln	6	5.99	5.22
R Arg	2	2.43	1.74
S Ser	7	4.75	6.09
T Thr	9	7.09	7.83
V Val	15	11.58	13.04
W Trp	0	0.00	0.00
Y Tyr	2	2.54	1.74
B Asx	0	0.00	0.00
Z Glx	0	0.00	0.00
X Xxx	0	0.00	0.00
. Ter	0	0.00	0.00

Fig. 4.10b

1 GGCACGAGCAAAAATGAAACTCCTCTTGTGCTTTGCTTCGCCGCC
 M K L L L C F A F A A
 47 ATCGTCATCGGAGCTCAGGCTCTCACCGATGAACAGATACAGAAA
 I V I G A Q A L T D E Q I Q K ↑
 92 AGGAACAAGATCAGCAAAGAATGCCAGCAGGAGTCCGGAGTGTCC
 R N K I S K E C Q Q E S G V S
 137 CAAGAGACGATCGACAAAGTCCGCACAGGTGTCTTGGTCGACGAT
 Q E T I D K V R T G V L V D D
 182 CCCAAATGAAGAACGACGTCTGCTCTGCTTCTCGAAGAGAAACTGGA
 P K M K H V L C F S K R T G
 226 GTGGCAACCGAAGCAGCCGGAGACACCAATGTGGAGGTACTCAAAGCC
 V A T E A G D T N V E V L K A
 271 AAGCTGAAGCATGTGGCCAGCGACGAAGAAGTGGACAAAGATCGTG
 K L K H V A S D E E V D K I V
 316 CAGAACGTGCGTGGTCAAGAACGCCACACCAAGAGGAAACGGCTTAT
 Q K C V V K K A T P E E T A Y
 361 GACACCTTCAAGTGTATTACGACAGTAACCTGATTCTCTCCT
 D T F K V I Y D S K P D F S P
 406 ATTGATTAATTGTTGTATTGACTGAATTGACAAATAAAGGT
 I D * polyadenylation signal
 451 ACTATCGTTATGAAAAAAAAAAAAAA

Fig. 4.11a

Predicted Amino Acid

Composition of 3-9

Analysis	Whole Protein
Molecular Weight	12871.80 m.w.
Length	115
1 microgram =	77.689 pMoles
Molar Extinction coefficient	3040±5%
1 A(280) =	4.23 mg/ml
Isoelectric Point	7.11
Charge at pH 7	0.13

Whole Protein Composition Analysis

Amino Acid(s)	Number count	% by weight	% by frequency
Charged (RKHYCDE)	48	47.31	41.74
Acidic (DE)	20	18.86	17.39
Basic (KR)	20	20.57	17.39
Polar (NCQSTY)	30	25.29	26.09
Hydrophobic (AILFWV)	34	27.20	29.57
A Ala	6	3.31	5.22
C Cys	4	3.21	3.48
D Asp	11	9.84	9.57
E Glu	9	9.03	7.83
F Phe	3	3.43	2.61
G Gly	4	1.77	3.48
H His	2	2.13	1.74
I Ile	6	5.28	5.22
K Lys	17	16.93	14.78
L Leu	5	4.40	4.35
M Met	1	1.02	0.87
N Asn	2	1.77	1.74
P Pro	4	3.02	3.48
Q Gln	6	5.97	5.22
R Arg	3	3.64	2.61
S Ser	7	4.74	6.09
T Thr	9	7.07	7.83
V Val	14	10.78	12.17
W Trp	0	0.00	0.00
Y Tyr	2	2.54	1.74
B Asx	0	0.00	0.00
Z Glx	0	0.00	0.00
X Xxx	0	0.00	0.00
. Ter	0	0.00	0.00

Fig. 4.11b

1000 900 800 700 600 500 400 300 200 100

1 GGCACGGAGCAAAAATGAAACTCCTCTTGTGCTTTCGCGTTGCCGCC
 M K L L L C F A F A A

47 ATCGTCATCGGAGCTCAGGCTCTCACCGACGAACAGATAACAGAAA
 I V I G A Q A L T D E Q I Q K

92 AGGAACACAAGATCAGCAAAGAGTGCCAGCAGGTGTCCGGAGTGTCC
 R N K I S K E C Q Q E S G V S

137 CAAGAGAGACGATCGACAAAGTCCGCACAGGTGTCTTGGTCGACGAT
 Q E T I D K V R T G V L V D D

182 CCCAAAATGAAGAACGACGTCCCTGCTTCTCGAAGAAAAACTGGA
 P K M K K H V L C F S K R T G

226 GTGGCAACCGAACGCCGGAGACACCAATGTGGAGGTACTCAAAGCC
 V A T E A G D T N V E V L K A

271 AAGCTGAAGCATGTGGCCAGCGACGAAGAAGTGGACAAGATCGTG
 K L K H V A S D E E V D K I V

316 CAGAAAGTGCCTGGTCAAGAACGGCCACACCAAGAGGAACGGCTTAT
 Q K C V V K K A T P E E T A Y

361 GACACCTTCAAGTGTATTTACGACAGTAAACCTGATTTCTCCT
 D T F K V I Y D S K P D F S P

406 ATTGATTAATTGTTTGTATTTGGCTGAATTTGACAAATAAGGT
 I D

polyadenylation signal

451 ACTATCGTTATGTAAAAAAAAAAAAAA

poly (A) tail

Fig. 4.12a

Predicted Amino Acid

Composition of 7-5

Analysis	Whole Protein
Molecular Weight	12843.80 m.w.
Length	115
1 microgram =	77.859 pMoles
Molar Extinction coefficient	3040±5%
1 A(280) =	4.22 mg/ml
Isoelectric Point	7.11
Charge at pH 7	0.13

Whole Protein Composition Analysis

Amino Acid(s)	Number count	% by weight	% by frequency
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Polar (NCQSTY)	30	25.35	26.09
Hydrophobic (AILFWV)	34	27.26	29.57
A Ala	6	3.32	5.22
C Cys	4	3.21	3.48
D Asp	11	9.86	9.57
E Glu	9	9.05	7.83
F Phe	3	3.44	2.61
G Gly	4	1.78	3.48
H His	2	2.14	1.74
I Ile	6	5.29	5.22
K Lys	18	17.97	15.65
L Leu	5	4.41	4.35
M Met	1	1.02	0.87
N Asn	2	1.78	1.74
P Pro	4	3.02	3.48
Q Gln	6	5.98	5.22
R Arg	2	2.43	1.74
S Ser	7	4.75	6.09
T Thr	9	7.08	7.83
V Val	14	10.80	12.17
W Trp	0	0.00	0.00
Y Tyr	2	2.54	1.74
B Asx	0	0.00	0.00
Z Glx	0	0.00	0.00
X Xxx	0	0.00	0.00
. Ter	0	0.00	0.00

Fig. 4.12b

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Fig 4.13

<pre> 2-2 M K L L C F A F A A V G A Q A L T D E Q I Q K R N K I S K E C Q Q V S G V S Q E T D K V R T G V L V 2-3 M K L L C F A F A A V G A Q A L T D E Q I Q K R N K I S K E C Q Q V S G V S Q E T D K V R T G V L V 3-4 M K L L C F A F A A V G A Q A L T D E Q I Q K R N K I S K E C Q Q V S G V S Q E T D K V R T G V L V 3-9 M K L L C F A F A A V G A Q A L T D E Q I Q K R N K I S K E C Q Q V S G V S Q E T D K V R T G V L V 7-5 M K L L C F A F A A V G A Q A L T D E Q I Q K R N K I S K E C Q Q V S G V S Q E T D K V R T G V L V </pre>	<pre> 2-2 D D P K M K K H V L C F S K K T G V A T E A G D T N V E V L K A K L K H V A S D E E V D K I V Q K C V V K K 2-3 D D P K M K K H V L C F S K K T G V A T E A G D T N V E V L K A K L K H V A S D E E V D K I V Q K C V V K K 3-4 D D P K M K K H V L C F S K K T G V A T E A G D T N V E V L K A K L K H V A S D E E V D K I V Q K C V V K K 3-9 D D P K M K K H V L C F S K K T G V A T E A G D T N V E V L K A K L K H V A S D E E V D K I V Q K C V V K K 7-5 D D P K M K K H V L C F S K K T G V A T E A G D T N V E V L K A K L K H V A S D E E V D K I V Q K C V V K K </pre>	<pre> 2-2 A T P E E E T A Y D T F K C I Y D S K P D F S P I D * 2-3 A T P E E E T A Y D T F K C I Y D S K P D F S P I D * 3-4 A T P E E E T A Y D T F K C I Y D S K P D F S P I D * 3-9 A T P E E E T A Y D T F K C I Y D S K P D F S P I D * 7-5 A T P E E E T A Y D T F K C I Y D S K P D F S P I D * </pre>
-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------	-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------	-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------

Fig. 4.14

MW (kDa)	AA (#)	(百分之 mole)												% most hydrophilic							
		Cys	Pro	Phe	Ile	Val	Met	Leu	% most hydrophobic	Gly	Ala	Tyr	His	Trp	Asx	Glx	Arg	Lys	Ser	Thr	
Tm 12.86	117	3.2	3.0	3.4	4.4	8.5	2.0	4.4	28.9	3.1	3.9	3.8	3.2	ND	10.7	15.0	3.6	14.9	6.8	6.3	
Tm 13.17	116	3.13	2.21	4.47	5.16	10.5	1.0	4.3	29.11	1.73	3.24	0	0	1.41	0	0	7.12	15.6	3.31	6.14	
2-2	12.84	115	3.21	3.02	3.44	5.29	10.8	1.02	4.41	27.26	1.78	3.32	2.54	2.14	0	0	0	2.43	18.0	4.75	7.08
2-3	12.84	115	3.21	3.02	3.44	5.29	10.8	1.02	4.41	27.26	1.78	3.32	2.54	2.14	0	0	0	2.43	18.0	4.75	7.08
3-4	12.84	115	2.41	3.02	3.44	5.29	11.6	1.02	4.41	28.04	1.78	3.32	2.54	2.14	0	0	0	2.43	18.0	4.75	7.09
3-9	12.87	115	3.21	3.02	3.43	5.28	10.8	1.02	4.40	27.20	1.77	3.31	2.54	2.13	0	0	0	3.64	16.9	4.74	7.07
7-5	12.84	115	3.21	3.02	3.44	5.29	10.8	1.02	4.41	27.26	1.78	3.32	2.54	2.14	0	0	0	2.43	18.0	4.75	7.08

Fig. 4.15

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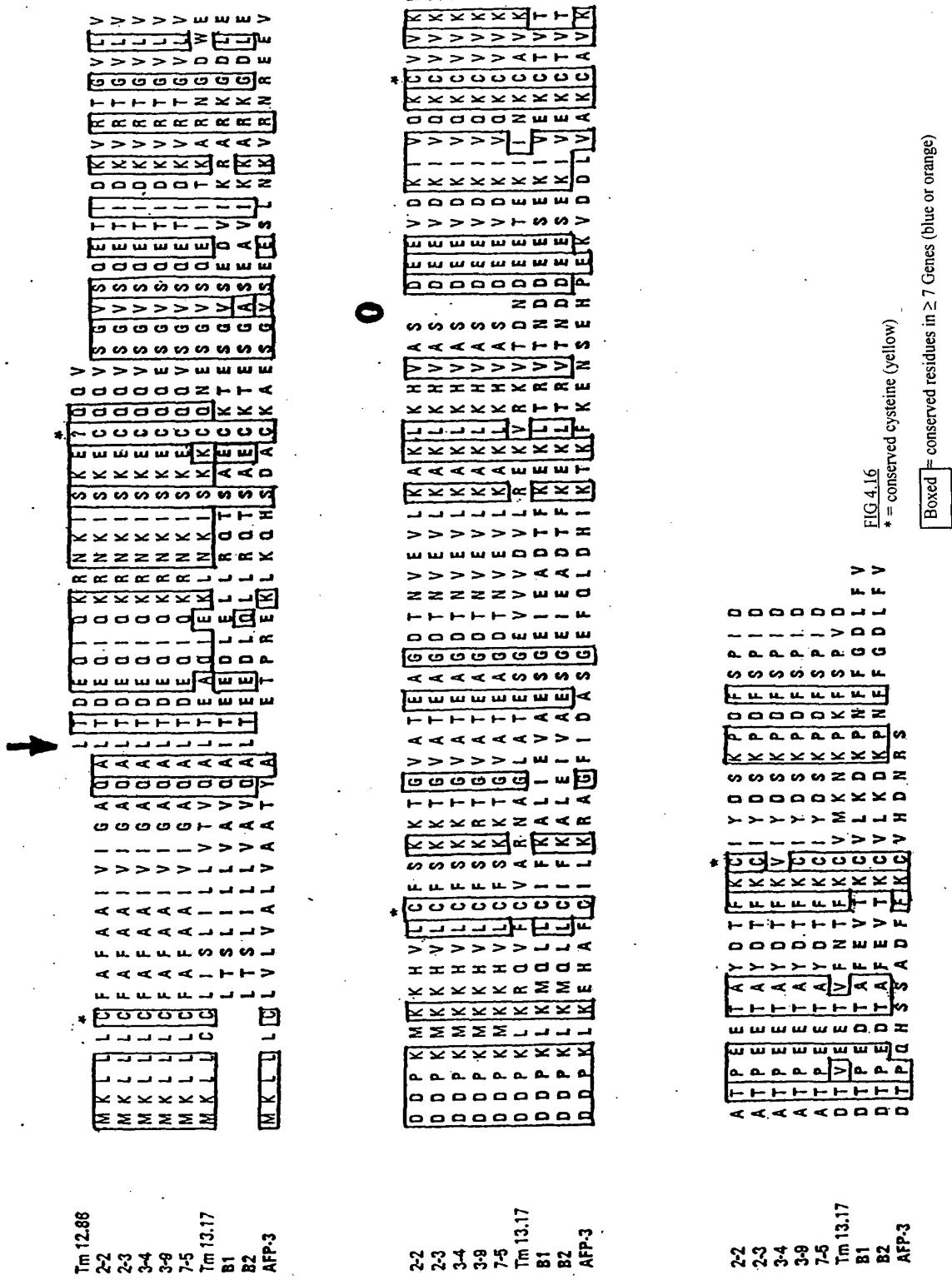


Fig. 4.16

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→ = conserved cysteine (yellow); Boxed = conserved residues in Genes (red)

Tm 13.17	34	R N G D
2 ²	34	R T G D
B1 prot	34	R K G D
B2 prot	34	R K G D
Pbp_Antp	34	R K G D
Pbp_Manso	34	R K G D
Qbp1_Manso	34	R K G D
Qbp2_Manso	34	R K G D
Pbpas-E_Drone	40	D L Y N F W K D
Pbpas-F_Drone	40	D F A N F W V E
Pbpmp1_Drone	40	F H F W R E D
Pbpmp2_Drone	36	S D G C
	35	V K N R
	36	M S H D

Fig. 4.17

PROTEIN SEQUENCE ALIGNMENT

Tm 12.86

2-2	M[K][L]LLC[F]FAAIVIGAAQAL	SKE[Q]QVSSGVSQETD
2-3	M[K][L]LLC[F]FAAIVIGAAQAL	SKE[Q]QVSSGVSQETD
3-4	M[K][L]LLC[F]FAAIVIGAAQAL	SKE[Q]QVSSGVSQETD
3-8	M[K][L]LLC[F]FAAIVIGAAQAL	SKE[Q]QVSSGVSQETD
7-5	M[K][L]LLC[F]FAAIVIGAAQAL	SKE[Q]QVSSGVSQETD
Tm 13.17	M[K][L]LLC[G]FASLLVTVQAL	SKE[Q]QVSSGVSQETD
B1	LTSLLVAVQALTEED	D[E]VQKCVV
B2	LTSLLVVALVAA[T]Y	D[E]VQKCVV
AFP-3	M[K][L]LLVVALVAA[T]Y	D[E]VQKCVV

Tm 13.17

2-2	DOPKMKKHVVL[G]FSKKTG	VATEAGDTNVEVLKAKLKHVAS
2-3	DOPKMKKHVVL[G]FSKKTG	VATEAGDTNVEVLKAKLKHVAS
3-4	DOPKMKKHVVL[G]FSKKTG	VATEAGDTNVEVLKAKLKHVAS
3-8	DOPKMKKHVVL[G]FSKKTG	VATEAGDTNVEVLKAKLKHVAS
7-5	DOPKMKKHVVL[G]FSKKTG	VATEAGDTNVEVLKAKLKHVAS
Tm 13.17	DOPKLKMQLLCIFKALEIVAE	SGEIVADTFKEKLTRVTNDDEESEKILV
B1	DOPKLKMQLLCIFKALEIVAE	SGEIVADTFKEKLTRVTNDDEESEKILV
B2	DOPKLKMQLLCIFKALEIVAE	SGEIVADTFKEKLTRVTNDDEESEKILV
AFP-3	DOPKLKMQLLCIFKALEIVAE	SGEIVADTFKEKLTRVTNDDEESEKILV

Tm 13.17

2-2	ATP[E]TAYDTF[K]CIVDSSKP	D
2-3	ATP[E]TAYDTF[K]CIVDSSKP	D
3-4	ATP[E]TAYDTF[K]CIVDSSKP	D
3-8	ATP[E]TAYDTF[K]CIVDSSKP	D
7-5	ATP[E]TAYDTF[K]CIVDSSKP	D
Tm 13.17	ATP[E]TAYDTF[K]CIVDSSKP	D
B1	ATP[E]TAYDTF[K]CIVDSSKP	D
B2	ATP[E]TAYDTF[K]CIVDSSKP	D
AFP-3	ATP[E]TAYDTF[K]CIVDSSKP	D

FIG 4.18
* = conserved cysteine (yellow)
Boxed = conserved residues in Genes (red, green, orange or blue)

Fig. 4.18

NUCLEOTIDE SEQUENCES

		PERCENT SIMILARITY												
		1 ²⁻²	2 ²⁻³	3 ³⁻⁴	4 ³⁻⁹	5 ⁷⁻⁵	6 Tm13.17	7 B1	8 AFP-3	9 YL-1	10 DAFP-1A			
PERCENT DIVERGENCE	1	99.1	97.3	98.4	98.2	50.4	42	42.4	20.7	20.9	1	2-2		
	2	0.9	99	99	99	51.5	43.2	42.7	20.2	21.3	2	2-3		
3	1.6	60	98	98	50.3	42	42.1	20	20	3	3-4			
4	1.6	20	80	98.4	51.3	43.7	43.6	20.4	22	4	3-9			
5	1.8	25	75	45	50.4	42.5	42.9	22.2	23.1	5	7-5			
6	36.9	36.4	37.2	36.4	37.1	57.2	37.4	23.3	21	6	Tm 13.17			
7	39.9	41.2	41.7	40.7	41.2	30.2	39.3	21.9	22.1	7	B1			
8	41.8	42.4	43.1	42.3	42.3	45.3	49.4	21.8	23.1	8	AFP-3			
9	48.7	58.6	52	51.2	51.9	61.7	62.2	58.2	45.6	9	YL-1			
10	51.4	62.7	46.4	46.5	46.5	60	58.1	61.8	35.5	10	DAFP-1A			

AMINO ACID SEQUENCES

		PERCENT SIMILARITY												
		1 ²⁻²	2 ²⁻³	3 ³⁻⁴	4 ³⁻⁹	5 ⁷⁻⁵	6 Tm13.17	7 B1	8 AFP-3	9 YL-1	10 DAFP-1A			
PERCENT DIVERGENCE	1	100	99.1	99.1	100	51.3	37.4	35.2	11.6	12	1	2-2		
	2	0	99.1	99.1	100	51.3	37.4	35.2	11.6	12	2	2-3		
3	0.9	0.9	98.3	99.1	50.4	36.5	34.3	11.6	12	3	3-4			
4	0.9	0.9	1.7	99.1	51.3	37.4	36.1	10.7	12	4	3-9			
5	0	0	0.9	0.09	51.3	37.4	35.2	11.6	12	5	7-5			
6	46.1	46.1	47	46.1	46.1	47.4	39.8	13.4	13.9	6	Tm 13.17			
7	59.1	59.1	60	59.1	59.1	51.7	37	11.6	11.1	7	B1			
8	61.7	61.7	62.6	60.7	61.7	60.2	63	10.2	8.3	8	AFP-3			
9	86.7	86.7	85.7	85.7	85.7	86.8	84.2	87.5	55.6	9	YL-1			
10	88.3	88.3	86.4	84.5	85.4	89.1	82.7	90.5	40.2	10	DAFP-1A			

Fig. 4.19

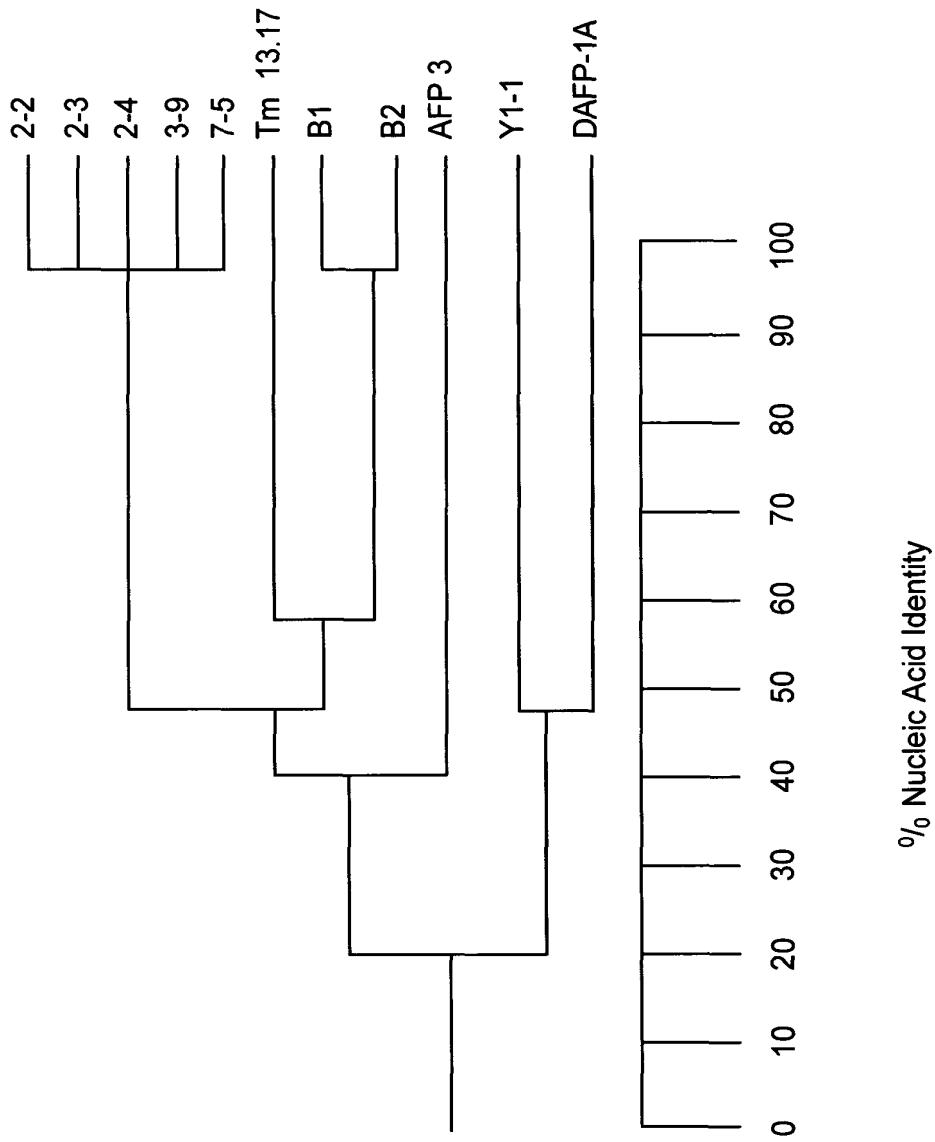


Fig. 4.20

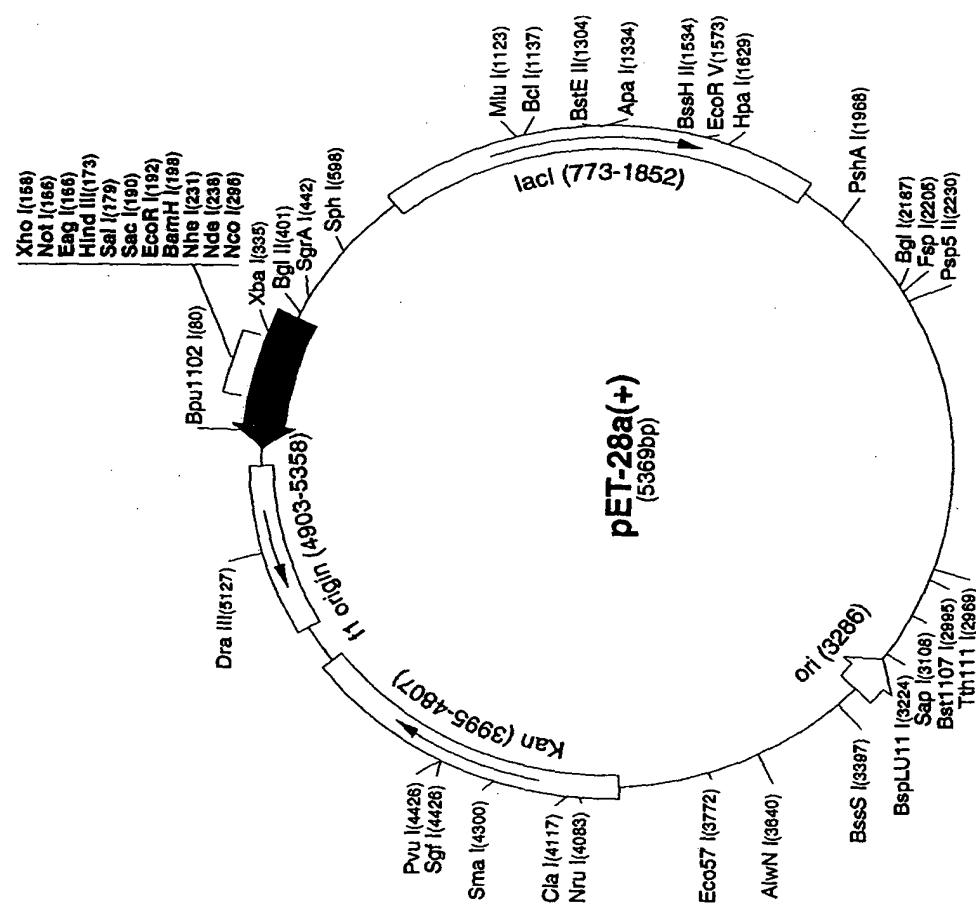


Fig. 5.0

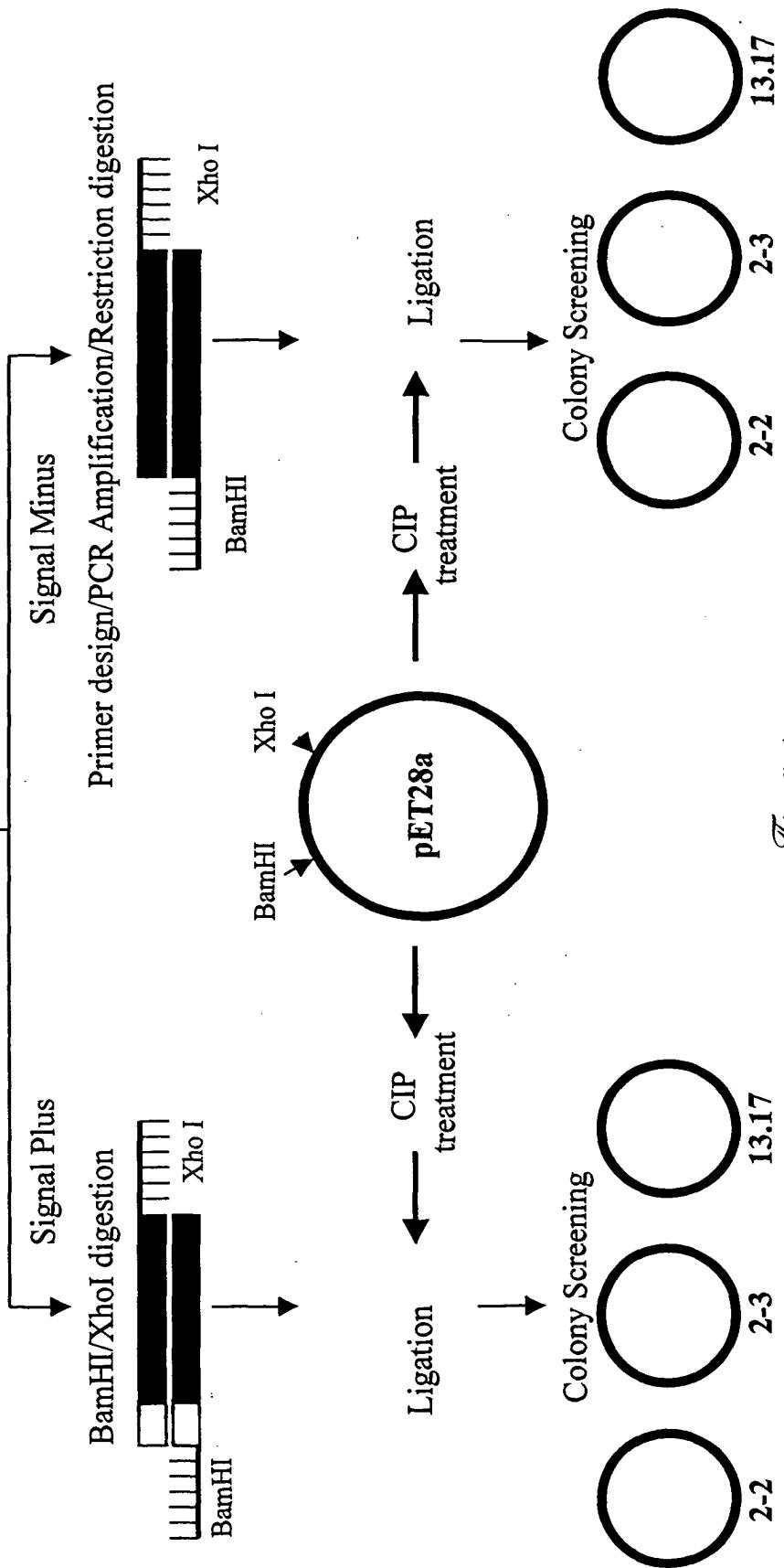
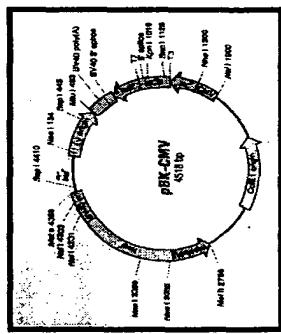


Fig. 5.1

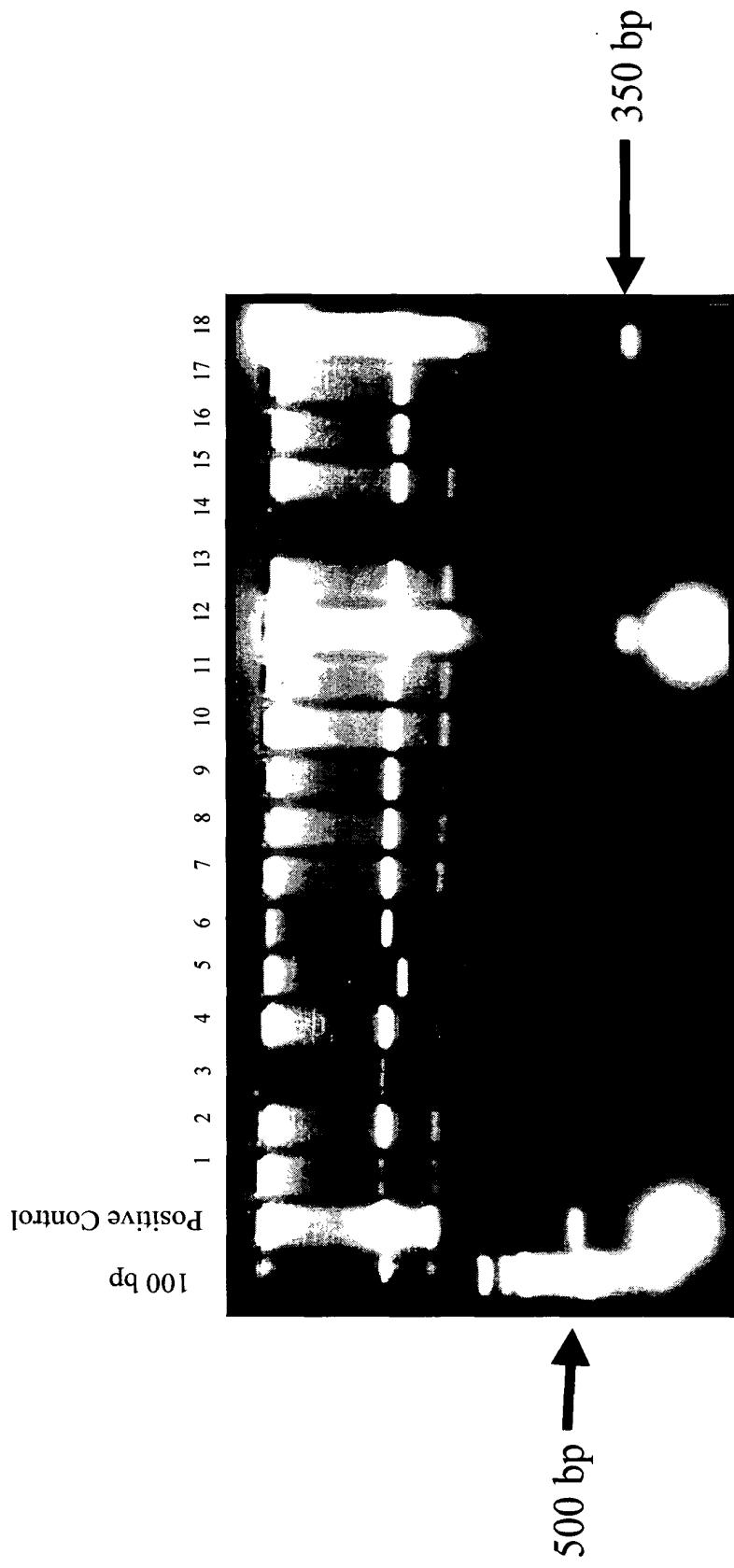


Fig. 5.2

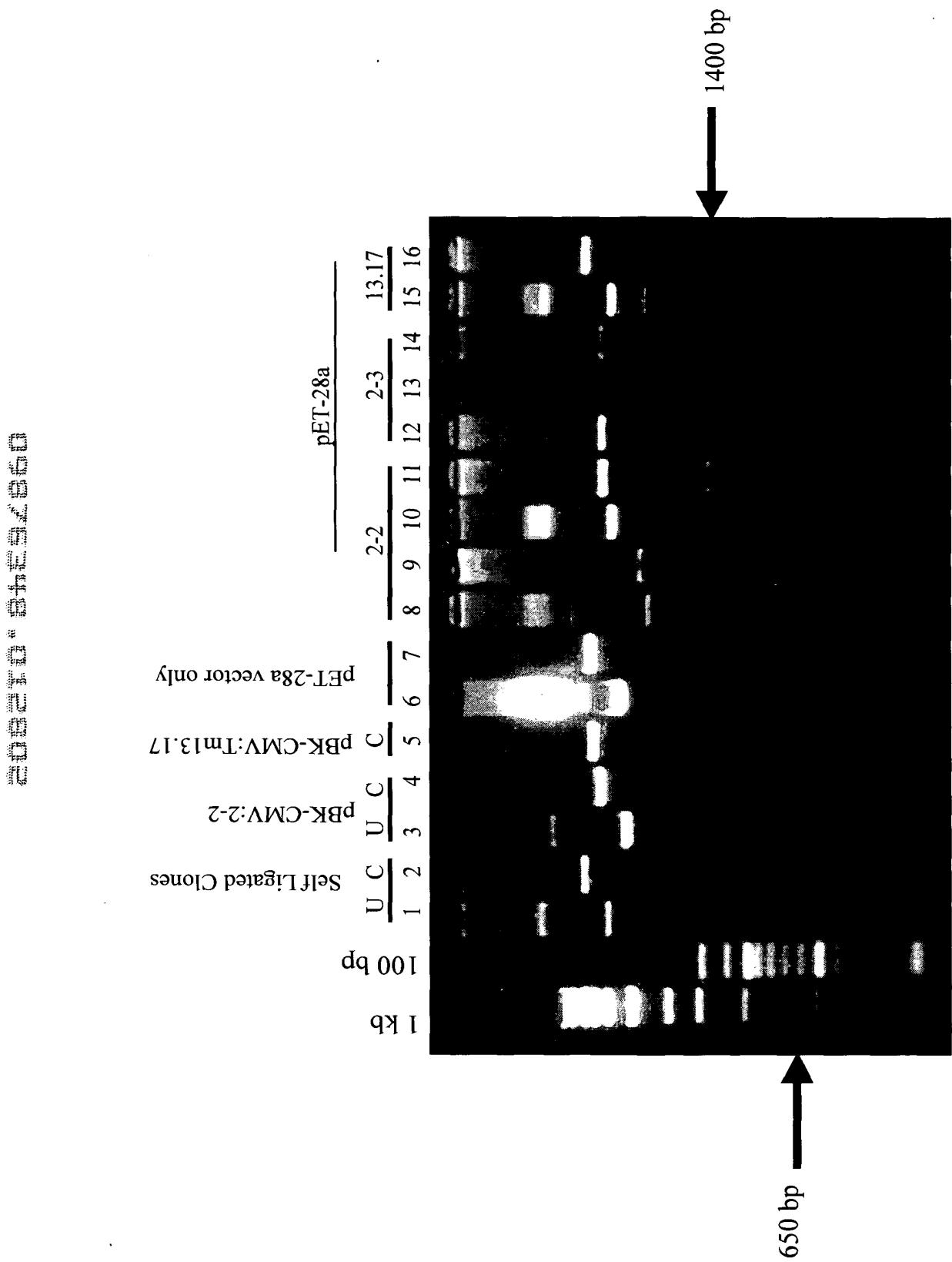
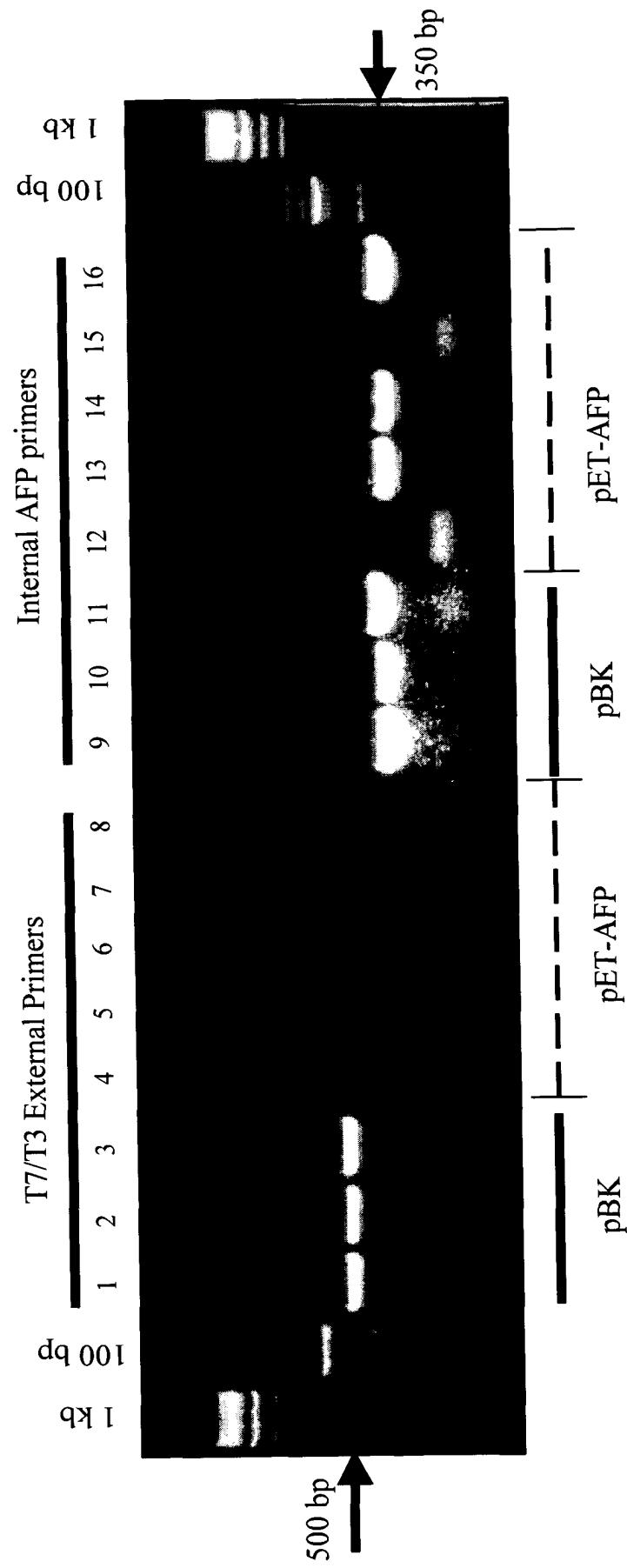


Fig. 5.3

Fig. 5.4



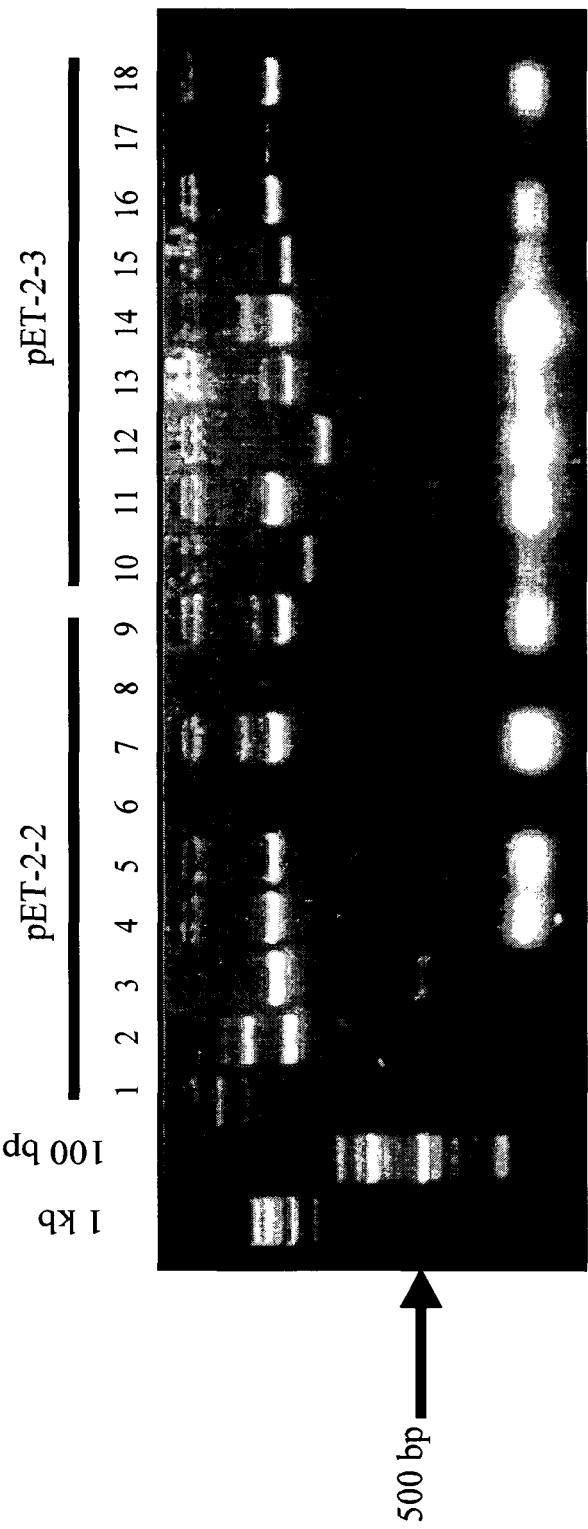


Fig. 5.5

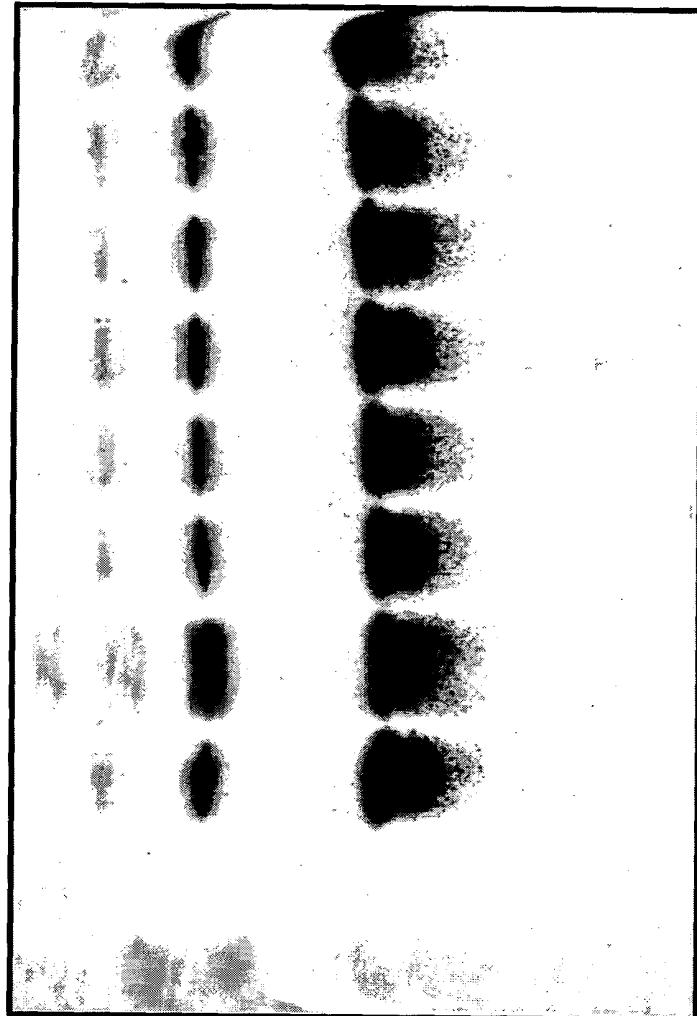
2-2 S+ 2-2 S- 2-3 S+ 2-3 S- 13.17 S+ 13.17 S-

Bacterial Protein lysate

Purified Tm12.86

Hemolymph

pET-28a Recombinant Histidine cleaved AFP



46 kD 29 kD 20 kD 14 kD 8 kD 3.5 kD

Fig. 5.6

His-tagged Clone 2.2 with signal sequence

His-tagged Clone 2.2 with signal sequence		
TTGTTAGCGG ATGGAATTCC CTCGTAGGGG ATAATTTGT TTACTTTAAG	50	
His-tag Start Codon		
AAGGAGATAT ACC ATG GGC AGC AGC CAT CAT CAT CAT CAC AGC	96	
Met Gly Ser Ser His His His His His His Ser		
-55	-50	
AGC GGC CTG GTG CCG CGC GGC AGC CAT ATG GCT AGC ATG ACT GGT	141	
Ser Gly Leu Val Pro Arg Gly Ser His Met Ala Ser Met Thr Gly		
-45	-40	-35
AFP Start Codon		
GGA CAG CAA ATG GGT CGC GGA TCC GAA TTC GCA CGA GCA AAA ATG	186	
Gly Gln Gln Met Gly Arg Gly Ser Glu Phe Ala Arg Ala Lys <u>Met</u>		
-30	-25	-20
AAA CTC CTC TTG TGC TTT GCG TTC GCC GCC ATC GTC ATC GGA GCT	231	
Lys Leu Leu Leu Cys Phe Ala Phe Ala Ala Ile Val Ile Gly Ala		
-15	-10	-5
N-terminal of mature AFP		
CAG GCT CTC ACC GAC GAA CAG ATA CAG AAA AGG AAC AAG ATC AGC	276	
<u>Gln</u> <u>Ala</u> Leu Thr Asp Glu Gln Ile Gln Lys Arg Asn Lys Ile Ser		
1	5	10
AAA GAA TGC CAG CAG GTG TCC GGA GTG TCC CAA GAG ACG ATC GAC	321	
Lys Glu Cys Gln Gln Val Ser Gly Val Ser Gln Glu Thr Ile Asp		
15	20	25
AAA GTC CGC ACA GGT GTC TTG GTC GAT GAT CCC AAA ATG AAG AAG	366	
Lys Val Arg Thr Gly Val Leu Val Asp Asp Pro Lys Met Lys Lys		
30	35	40
CAC GTC CTC TGC TTC TCG AAG AAA ACT GGA GTG GCA ACC GAA GCC	411	
His Val Leu Cys Phe Ser Lys Lys Thr Gly Val Ala Thr Glu Ala		
45	50	55
GGA GAC ACC AAT GTG GAG GTA CTC AAA GCC AAG CTG AAG CAT GTG	456	
Gly Asp Thr Asn Val Glu Val Leu Lys Ala Lys Leu Lys His Val		
60	65	70
GCC AGC GAC GAA GAG GTG GAC AAG ATC GTG CAG AAG TGC GTG GTC	501	
Ala Ser Asp Glu Val Asp Lys Ile Val Gln Lys Cys Val Val		
75	80	85
AAG AAG GCC ACA CCA GAG GAA ACG GCT TAT GAC ACC TTC AAG TGT	546	
Lys Lys Ala Thr Pro Glu Glu Thr Ala Tyr Asp Thr Phe Lys Cys		
90	95	100
Stop Codon		
ATT TAC GAC AGT AAA CCT GAT TTC TCT CCT ATT GAT TAA TTGTTTGTA	595	
Ile Tyr Asp Ser Lys Pro Asp Phe Ser Pro Ile Asp *		
105	110	115
Polyadenylation signal Poly-A tail		
TTTGACTGAA TTTTGACAAT AAAGGTAATA TCGTTATGTA AAAAAAAAAA	645	
AAAAAAACTCG AGCACCAACCA CCACCAACAC TGAGAT		
681		

Fig. 5.7

His-tagged clone 2.2 without signal sequence		
TTGTTAGCGG ATGGAATTCC CTCGTAGGGG ATAATTTGT TTACTTAAG		50
His-tag Start Codon		
AAGGAGATAT ACC ATG GGC AGC AGC CAT CAT CAT CAT CAC AGC		96
Met Gly Ser Ser His His His His His His Ser		
-30		-25
AGC GGC CTG GTG CCG CGC GGC AGC CAT ATG GCT AGC ATG ACT GGT		141
Ser Gly Leu Val Pro Arg Gly Ser His Met Ala Ser Met Thr Gly		
-20		-10
N-terminal of mature AFP		
GGA CAG CAA ATG GGT CGC GGA TCC CTC ACC GAC GAA CAG ATA CAG		186
Gly Gln Gln Met Gly Arg Gly Ser Leu Thr Asp Glu Gln Ile Gln		
-5	1	5
AAA AGG AAC AAG ATC AGC AAA GAA TGC CAG CAG GTG TCC GGA GTG		231
Lys Arg Asn Lys Ile Ser Lys Glu Cys Gln Gln Val Ser Gly Val		
10	15	20
TCC CAA GAG ACG ATC GAC AAA GTC CGC ACA GGT GTC TTG GTC GAT		276
Ser Gln Glu Thr Ile Asp Lys Val Arg Thr Gly Val Leu Val Asp		
25	30	35
GAT CCC AAA ATG AAG AAG CAC GTC CTC TGC TTC TCG AAG AAA ACT		321
Asp Pro Lys Met Lys Lys His Val Leu Cys Phe Ser Lys Lys Thr		
40	45	50
GGA GTG GCA ACC GAA GCC GGA GAC ACC AAT GTG GAG GTA CTC AAA		366
Gly Val Ala Thr Glu Ala Gly Asp Thr Asn Val Glu Val Leu Lys		
55	60	65
GCC AAG CTG AAG CAT GTG GCC AGC GAC GAA GAG GTG GAC AAG ATC		411
Ala Lys Leu Lys His Val Ala Ser Asp Glu Glu Val Asp Lys Ile		
70	75	80
GTG CAG AAG TGC GTG GTC AAG AAG GCC ACA CCA GAG GAA ACG GCT		456
Val Gln Lys Cys Val Val Lys Lys Ala Thr Pro Glu Glu Thr Ala		
85	90	95
TAT GAC ACC TTC AAG TGT ATT TAC GAC AGT AAA CCT GAT TTC TCT		501
Tyr Asp Thr Phe Lys Cys Ile Tyr Asp Ser Lys Pro Asp Phe Ser		
100	105	110
Stop Codon		
CCT ATT GAT TAA CTCGAGCACC ACCACCA CCACGTGAGAT		543
Pro Ile Asp *		
115		

Fig. 5.8

卷之三

His-tag clone 2.3 with signal sequence	
TTGTTAGCGG ATGGAATTCC CTCGTAGGGG ATAATTTGT TTACTTTAAG	50
His-tag Start Codon	
AAGGAGATAT ACC ATG GGC AGC AGC CAT CAT CAT CAT CAC AGC Met Gly Ser Ser His His His His His His Ser	96
-55 -50	
AGC GGC CTG GTG CCG CGC GGC AGC CAT ATG GCT AGC ATG ACT GGT Ser Gly Leu Val Pro Arg Gly Ser His Met Ala Ser Met Thr Gly	141
-45 -40 -35	
GGA CAG CAA ATG GGT CGC GGA TCC GAA TTC GCA CGA GCA AAA ATG Gly Gln Gln Met Gly Arg Gly Ser Glu Phe Ala Arg Ala Lys Met	186
-30 -25 -20	
AAA CTC CTC TTG TGC TTT GCT TTC GCC GCC ATC GTC ATC GGA GCT Lys Leu Leu Leu Cys Phe Ala Phe Ala Ala Ile Val Ile Gly Ala	231
-15 -10 -5	
N-terminal of Mature AFP	
CAG GCT CTC ACC GAC GAA CAG ATA CAG AAA AGG AAC AAG AAG ATC AGC Gln Ala Leu Thr Asp Glu Gln Ile Gln Lys Arg Asn Lys Ile Ser	276
1 5 10	
AAA GAA TGC CAG CAG GTG TCC GGA GTG TCC CAA GAG ACG ACG ATC GAC Lys Glu Cys Gln Gln Val Ser Gly Val Ser Gln Glu Thr Ile Asp	321
15 20 25	
AAA GTC CGC ACA GGT GTC TTG GTC GAT GAT CCC AAA ATG AAG AAG Lys Val Arg Thr Gly Val Leu Val Asp Asp Pro Lys Met Lys Lys	366
30 35 40	
CAC GTC CTC TGC TTC TCG AAG AAA ACT GGA GTG GCA ACC GAA GCC His Val Leu Cys Phe Ser Lys Lys Thr Gly Val Ala Thr Glu Ala	411
45 50 55	
GGA GAC ACC AAT GTG GAG GTA CTC AAA GCC AAG CTG AAG CAT GTG Gly Asp Thr Asn Val Glu Val Leu Lys Ala Lys Leu Lys His Val	456
60 65 70	
GCC AGC GAC GAA GAA GTG GAC AAG ATC GTG CAG AAG TGC GTG GTC Ala Ser Asp Glu Glu Val Asp Lys Ile Val Gln Lys Cys Val Val	501
75 80 85	
AAG AAG GCC ACA CCA GAG GAA ACG GCT TAT GAC ACC TTC AAG TGT Lys Lys Ala Thr Pro Glu Glu Thr Ala Tyr Asp Thr Phe Lys Cys	546
90 95 100	
Stop Codon	
ATT TAC GAC AGT AAA CCT GAT TTC TCT CCT ATT GAT TAA TTGTTTGTA Ile Tyr Asp Ser Lys Pro Asp Phe Ser Pro Ile Asp *	595
105 110 115	
Polyadenylation signal Poly-A tail	
TTTGACTGAA TTTGACAAT AAAGGTACTA TCGTTATGAA AAAAAAAA AAAAAAAAACTC GAGCACCAACC ACCACCACTA CTGAGAT	645
682	

Fig. 5.9

His-tagged Clone 2.3 without signal sequence		
TTGTTAGCGG ATGGAATTCC CTCGTAGGGG ATAATTTTGT TTACTTTAAG		50
His-tag Start Codon		
AAGGAGATAT ACC ATG GGC AGC AGC CAT CAT CAT CAT CAC AGC		96
Met Gly Ser Ser His His His His His His Ser		
-30		-25
AGC GGC CTG GTG CCG CGC GGC AGC CAT ATG GCT AGC ATG ACT GGT		141
Ser Gly Leu Val Pro Arg Gly Ser His Met Ala Ser Met Thr Gly		
-20		-15
-10		
N-terminal of mature AFP		
GGA CAG CAA ATG GGT CGC GGA TCC CTC ACC GAC GAA CAG ATA CAG		186
Gly Gln Gln Met Gly Arg Gly Ser Leu Thr Asp Glu Gln Ile Gln		
-5		1
5		
AAA AGG AAC AAG ATC AGC AAA GAA TGC CAG CAG GTG TCC GGA GTG		231
Lys Arg Asn Lys Ile Ser Lys Glu Cys Gln Gln Val Ser Gly Val		
10		15
20		
TCC CAA GAG ACG ATC GAC AAA GTC CGC ACA GGT GTC TTG GTC GAT		276
Ser Gln Glu Thr Ile Asp Lys Val Arg Thr Gly Val Leu Val Asp		
25		30
35		
GAT CCC AAA ATG AAG AAG CAC GTC CTC TGC TTC TCG AAG AAA ACT		321
Asp Pro Lys Met Lys Lys His Val Leu Cys Phe Ser Lys Lys Thr		
40		45
50		
GGA GTG GCA ACC GAA GCC GGA GAC ACC AAT GTG GAG GTA CTC AAA		366
Gly Val Ala Thr Glu Ala Gly Asp Thr Asn Val Glu Val Leu Lys		
55		60
65		
GCC AAG CTG AAG CAT GTG GCC AGC GAC GAA GAA GTG GAC AAG ATC		411
Ala Lys Leu Lys His Val Ala Ser Asp Glu Glu Val Asp Lys Ile		
70		75
80		
GTG CAG AAG TGC GTG GTC AAG AAG GCC ACA CCA GAG GAA ACG GCT		456
Val Gln Lys Cys Val Val Lys Lys Ala Thr Pro Glu Glu Thr Ala		
85		90
95		
TAT GAC ACC TTC AAG TGT ATT TAC GAC AGT AAA CCT GAT TTC TCT		501
Tyr Asp Thr Phe Lys Cys Ile Tyr Asp Ser Lys Pro Asp Phe Ser		
100		105
110		
Stop Codon		
CCT ATT GAT TAA CTCGAGCACC ACCACCA CCACTGAGAT		543
Pro Ile Asp *		
115		

Fig. 5.10

His-tagged Tm 13.17 with signal sequence		
TTGTTAGCGG ATGGAATTCC CTCGTAGGGG ATAATTTTGT TTACTTTAAG	50	
His-tag Start Codon		
AAGGAGATAT ACC ATG GGC AGC AGC CAT CAT CAT CAT CAC AGC Met Gly Ser Ser His His His His His Ser	96	
-65 -60 -55		
AGC GGC CTG GTG CCG CGC GGC AGC CAT ATG GCT AGC ATG ACT GGT Ser Gly Leu Val Pro Arg Gly Ser His Met Ala Ser Met Thr Gly -50 -45 -40	141	
GGA CAG CAA ATG GGT CGC GGA TCC GAA TTC TGG ATC CAA AGA ATT Gly Gln Gln Met Gly Arg Gly Ser Glu Phe Trp Ile Gln Arg Ile -35 -30 -25	186	
AFP Start Codon		
CGG CAC GAG ACT ACT AAG ATG AAG TTG CTC TGT TGT CTA ATC TCC Arg His Glu Thr Thr Lys Met Lys Leu Leu Cys Cys Leu Ile Ser	231	
-20 -15 -10		
N-terminal of mature AFP		
CTC ATT CTG TTG GTC ACA GTT CAG GCC CTG ACC GAG GCA CAA ATT Leu Ile Leu Leu Val Thr Val Gln Ala Leu Thr Glu Ala Gln Ile	276	
-5 1 5		
GAG AAA CTG AAC AAG ATC AGC AAA AAA TGT CAA AAT GAA AGT GGA Glu Lys Leu Asn Lys Ile Ser Lys Lys Cys Gln Asn Glu Ser Gly 10 15 20	321	
GTG TCG CAA GAG ATC ATA ACC AAA GCT CGC AAC GGT GAC TGG GAG Val Ser Gln Glu Ile Ile Thr Lys Ala Arg Asn Gly Asp Trp Glu 25 30 35	366	
GAC GAT CCT AAA CTG AAA CGC CAA GTT TTT TGC GTG GCC AGG AAC Asp Asp Pro Lys Leu Lys Arg Gln Val Phe Cys Val Ala Arg Asn 40 45 50	411	
GCC GGT CTG GCC ACG GAA TCG GGA GAG GTG GTG GTC GAC GTG TTG Ala Gly Leu Ala Thr Glu Ser Gly Glu Val Val Val Asp Val Leu 55 60 65	456	
AGG GAG AAG GTG AGG AAG GTC ACT GAC AAC GAC GAA GAA ACT GAG Arg Glu Lys Val Arg Lys Val Thr Asp Asn Asp Glu Glu Thr Glu 70 75 80	501	
AAA ATC ATC AAT AAG TGC GCC GTC AAG AGA GAT ACT GTT GAA GAG Lys Ile Ile Asn Lys Cys Ala Val Lys Arg Asp Thr Val Glu Glu 85 90 95	546	
ACG GTG TTC AAT ACT TTC AAA TGT GTC ATG AAA AAC AAG CCA AAG Thr Val Phe Asn Thr Phe Lys Cys Val Met Lys Asn Lys Pro Lys 100 105 110	595	
Stop Codon		
TTC TCA CCA GTT GAT TGA ACCACCACGA CTAGTAGATG GTTCAAATGG Phe Ser Pro Val Asp *	643	
115		
Polyadenylation signal Poly-A tail		
TGTGCTTAC ATATAAAA <u>AT</u> AAAGTGTTC TGATGTAAAA AAAAAAAAAA	693	
AAAAAAAAAA AACTCGAGAG TATTCTAGAG CGGCCGCGGG CCCATCGTT	743	
TCCACCCCTC GAGCACCAACC ACCACCACTA CTGAGAT	777	

Fig. 5.11

His-tagged Tm 13.17 without signal sequence		
TTGTTAGCGG ATGGAATTCC CTCGTAGGGG ATAATTTGT TTACTTTAAG		50
His-tag Start Codon		
AAGGAGATAT ACC ATG GGC AGC AGC CAT CAT CAT CAT CAC AGC		96
Met Gly Ser Ser His His His His His His Ser		
-30	-25	
AGC GGC CTG GTG CCG CGC GGC AGC CAT ATG GCT AGC ATG ACT GGT		141
Ser Gly Leu Val Pro Arg Gly Ser His Met Ala Ser Met Thr Gly		
-20	-15	-10
N-terminal of mature AFP		
GGA CAG CAA ATG GGT CGC GGC CTG ACC GAG GCA CAA ATT GAG AAA		186
Gly Gln Gln Met Gly Arg Gly Leu Thr Glu Ala Gln Ile Glu Lys		
-5	1	5
CTG AAC AAG ATC AGC AAA AAA TGT CAA AAT GAA AGT GGA GTG TCG		231
Leu Asn Lys Ile Ser Lys Lys Cys Gln Asn Glu Ser Gly Val Ser		
10	15	20
CAA GAG ATC ATA ACC AAA GCT CGC AAC GGT GAC TGG GAG GAC GAT		276
Gln Glu Ile Ile Thr Lys Ala Arg Asn Gly Asp Trp Glu Asp Asp		
25	30	35
CCT AAA CTG AAA CGC CAA GTT TTT TGC GTG GCC AGG AAC GCC GGT		321
Pro Lys Leu Lys Arg Gln Val Phe Cys Val Ala Arg Asn Ala Gly		
40	45	50
CTG GCC ACG GAA TCG GGA GAG GTG GTG GTC GAC GTG TTG AGG GAG		366
Leu Ala Thr Glu Ser Gly Glu Val Val Val Asp Val Leu Arg Glu		
55	60	65
AAG GTG AGG AAG GTC ACT GAC AAC GAC GAA GAA ACT GAG AAA ATC		411
Lys Val Arg Lys Val Thr Asp Asn Asp Glu Glu Thr Glu Lys Ile		
70	75	80
ATC AAT AAG TGC GCC GTC AAG AGA GAT ACT GTT GAA GAG ACG GTG		456
Ile Asn Lys Cys Ala Val Lys Arg Asp Thr Val Glu Glu Thr Val		
85	90	95
TTC AAT ACT TTC AAA TGT GTC ATG AAA AAC AAG CCA AAG TTC TCA		501
Phe Asn Thr Phe Lys Cys Val Met Lys Asn Lys Pro Lys Phe Ser		
100	105	110
Stop Codon		
CCA GTT GAT TGA CTCGAGCACCC ACCACCACCA CCACTGAGAT		543
Pro Val Asp *		
115		

Fig. 5.12

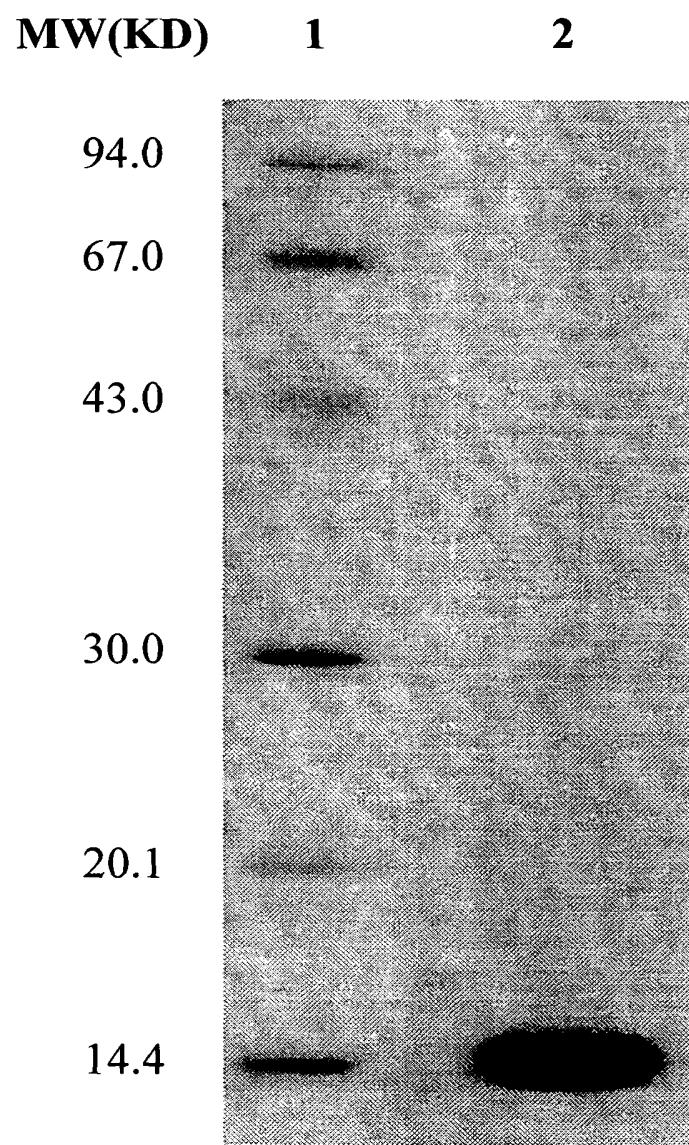


Fig. 6.0

1 2 3 MW(kb)

34.8

29.4

20.9

7.40



Fig. 6.1



Fig. 6.2

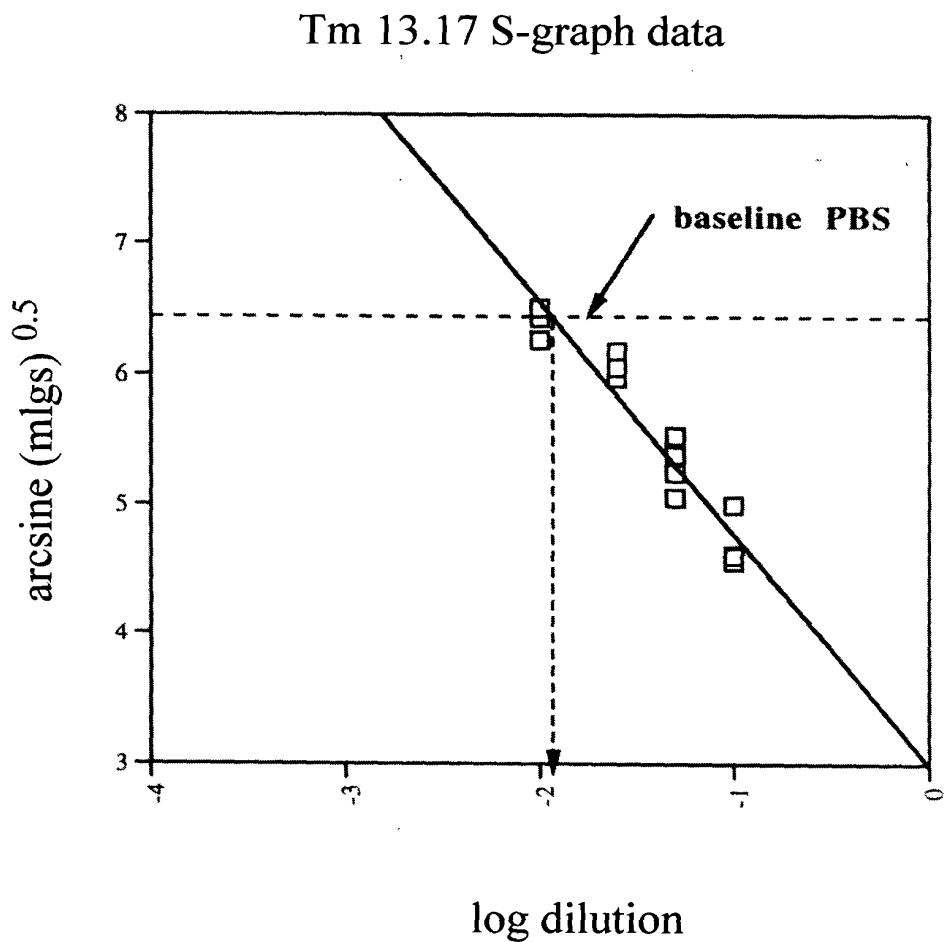


Fig. 6.3

ONE LETTER	NAME	THREE LETTER	CHEMICAL CLASS	HYDROPHOBICITY	Reactivity
A	Alanine	Ala	aliphatic	mod. hydrophobic	low
B	Asp or Asn	Asx			
C	Cysteine	Cys	sulphydryl	hydrophobic	high
D	Aspartic Acid	Asp	acidic	highly hydrophilic	high
E	Glutamic Acid	Glu	acidic	highly hydrophilic	high
F	Phenylalanine	Phe	aromatic	highly hydrophobic	low
G	Glycine	Gly	aliphatic	mod. hydrophobic	low
H	Histidine	His	basic, imidazole	highly hydrophilic	high
I	Isoleucine	Ile	aliphatic	hydrophobic	low
J					
K	Lysine	Lys	basic	highly hydrophilic	high
L	Leucine	Leu	aliphatic	hydrophobic	low
M	Methionine	Met	sulphydryl	hydrophobic	low
N	Asparagine	Asn	amide, acidic derived	hydrophilic	high
O					
P	Proline	Pro	aliphatic, cyclic, imino	mod. hydrophilic	low
Q	Glutamine	Gln	amide, acidic derived	hydrophilic	high
R	Arginine	Arg	basic	highly hydrophilic	high
S	Serine	Ser	aliphatic hydroxyl	hydrophilic	high
T	Threonine	Thr	aliphatic hydroxyl	hydrophilic	high
U					
V	Valine	Val	aliphatic	hydrophobic	low
W	Tryptophan	Trp	aromatic	highly hydrophobic	low
X					
Y	Tyrosine	Tyr	aromatic	mod. hydrophilic	high
Z	Glu or Gln	Glx			
		ACD	Any Acidic		
		ALP	Any Aliphatic		
		ALH	Any Aliphatic Hydroxyl		
		ARO	Any Aromatic		
		BAS	Any Basic		
		HY-	Hydrophobic		
		HY+	Hydrophilic		

Fig. 7.1

Sequence Alignment

Position	Tm12.84-2.2	Tm12.84-2.3	Tm12.84-3.4	Tm12.84-3.9	Tm12.84-7.5	Consensus of Tm12.84	Tm13.17	Consensus with Tm13.17	B1	Consensus with B1	AFP-3	Consensus with AFP-3
1	A	A	A	A	A	A	A	A	A?	A	CAG	NNNNNAAAR+
2	C	G	G	G	G	C	G	N	T?	N	ATC	NNNNNAAAR+
3	G	A	A	A	A	G	A	R	G?	R	CCG	NNNNNAAAR+
4	A	G	G	G	G	A	G	N		N	AAAG	NNNNNAAAR+
5	G	C	C	C	C	G	A	R		R	ATG	NNNNNAAAR+
6	A	A	A	A	A	A	A	A		A	AAAG	NNNNNAAAR+
7	A	A	A	A	A	A	A	A		T	ATGAA	NNNNNAAAR+
8	A	A	A	A	A	A	A	A		G	AGCT	NNNNNAAAR+
9	A	A	A	A	A	A	A	A		C	CTCT	NNNNNAAAR+
10	A	A	A	A	A	A	A	A		T	CTCT	NNNNNAAAR+
11	A	A	A	A	A	A	A	A		G	CTG	NNNNNAAAR+
12	A	A	A	A	A	A	A	A		T	CTT	NNNNNAAAR+
13	A	T	G	A	A	A	T	A		G	GT	NNNNNAAAR+
14	T	G	A	A	A	A	G	A		C	TC	NNNNNAAAR+
15	G	A	A	A	A	A	A	A		T	CT	NNNNNAAAR+
16	A	A	A	A	A	A	A	A		G	CT	NNNNNAAAR+
17	A	A	A	A	A	A	A	A		C	CG	NNNNNAAAR+
18	A	A	A	A	A	A	A	A		G	CG	NNNNNAAAR+
19	A	A	A	A	A	A	A	A		C	GG	NNNNNAAAR+
20	A	A	A	A	A	A	A	A		C	GC	NNNNNAAAR+
21	A	A	A	A	A	A	A	A		G	AC	NNNNNAAAR+
22	A	A	A	A	A	A	A	A		A	AG	NNNNNAAAR+
23	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
24	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
25	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
26	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
27	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
28	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
29	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
30	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
31	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
32	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
33	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
34	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
35	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
36	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
37	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
38	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
39	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
40	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
41	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
42	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
43	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
44	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
45	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
46	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
47	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
48	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
49	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
50	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
51	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
52	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
53	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
54	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
55	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
56	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
57	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
58	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
59	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
60	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
61	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
62	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
63	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
64	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
65	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
66	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
67	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
68	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
69	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
70	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
71	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
72	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
73	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
74	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
75	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
76	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
77	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
78	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
79	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
80	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
81	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
82	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
83	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
84	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
85	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
86	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
87	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
88	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
89	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
90	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
91	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
92	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
93	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
94	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
95	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
96	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
97	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
98	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
99	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
100	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
101	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
102	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
103	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
104	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
105	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
106	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
107	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
108	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
109	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
110	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
111	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
112	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
113	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
114	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
115	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
116	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+
117	A	A	A	A	A	A	A	A		N	AG	NNNNNAAAR+

Fig. 7.2

Position	Tm12.84-2.2	Tm12.84-2.3	Tm12.84-3.4	Tm12.84-3.9	Tm12.84-7.5	Consensus of	Tm13.17	Consensus with	B1	Consensus with	AFP-3	Consensus with
116	C	C	C	C	C	C	A	N	G	N	G	N
119	A	A	A	A	A	A	A	N	R	N	R	N
120	G	G	G	G	G	G	T	N	T	T	T	N
121	T	T	T	T	T	T	A	A	A	A	C	A
122	G	G	G	G	G	G	T	G	G	G	G	G
123	T	T	T	T	T	T	G	A	T	T	T	T
124	T	T	T	T	T	T	G	G	G	G	G	G
125	C	C	C	C	C	C	C	G	G	G	G	G
126	C	C	C	C	C	C	G	G	G	G	G	G
127	G	G	G	G	G	G	G	G	G	G	G	G
128	G	G	G	G	G	G	G	G	G	G	G	G
129	A	A	A	A	A	A	A	A	A	A	A	A
130	G	G	G	G	G	G	G	G	G	G	G	G
131	T	T	T	T	T	T	T	T	T	T	T	T
132	G	G	G	G	G	G	G	G	G	G	G	G
133	T	T	T	T	T	T	T	T	T	T	T	T
134	C	C	C	C	C	C	C	C	C	C	C	C
135	C	C	C	C	C	C	C	C	C	C	C	C
136	C	C	C	C	C	C	C	C	C	C	C	C
137	A	A	A	A	A	A	A	A	A	A	A	A
138	A	A	A	A	A	A	A	A	A	A	A	A
139	G	G	G	G	G	G	G	G	G	G	G	G
140	A	A	A	A	A	A	A	A	A	A	A	A
141	G	G	G	G	G	G	G	G	G	G	G	G
142	A	A	A	A	A	A	A	A	A	A	A	A
143	C	C	C	C	C	C	C	C	C	C	C	C
144	G	G	G	G	G	G	G	G	G	G	G	G
145	A	A	A	A	A	A	A	A	A	A	A	A
146	T	T	T	T	T	T	T	T	T	T	T	T
147	C	C	C	C	C	C	C	C	C	C	C	C
148	A	A	A	A	A	A	A	A	A	A	A	A
149	C	C	C	C	C	C	C	C	C	C	C	C
150	A	A	A	A	A	A	A	A	A	A	A	A
151	A	A	A	A	A	A	A	A	A	A	A	A
152	A	A	A	A	A	A	A	A	A	A	A	A
153	A	A	A	A	A	A	A	A	A	A	A	A
154	G	G	G	G	G	G	G	G	G	G	G	G
155	T	T	T	T	T	T	T	T	T	T	T	T
156	C	C	C	C	C	C	C	C	C	C	C	C
157	G	G	G	G	G	G	G	G	G	G	G	G
158	A	A	A	A	A	A	A	A	A	A	A	A
159	A	A	A	A	A	A	A	A	A	A	A	A
160	A	A	A	A	A	A	A	A	A	A	A	A
161	A	A	A	A	A	A	A	A	A	A	A	A
162	A	A	A	A	A	A	A	A	A	A	A	A
163	G	G	G	G	G	G	G	G	G	G	G	G
164	T	T	T	T	T	T	T	T	T	T	T	T
165	C	C	C	C	C	C	C	C	C	C	C	C
166	G	G	G	G	G	G	G	G	G	G	G	G
167	T	T	T	T	T	T	T	T	T	T	T	T
168	C	C	C	C	C	C	C	C	C	C	C	C
169	T	T	T	T	T	T	T	T	T	T	T	T
170	G	G	G	G	G	G	G	G	G	G	G	G
171	A	A	A	A	A	A	A	A	A	A	A	A
172	T	T	T	T	T	T	T	T	T	T	T	T
173	G	G	G	G	G	G	G	G	G	G	G	G
174	A	A	A	A	A	A	A	A	A	A	A	A
175	C	C	C	C	C	C	C	C	C	C	C	C
176	T	T	T	T	T	T	T	T	T	T	T	T
177	G	G	G	G	G	G	G	G	G	G	G	G
178	A	A	A	A	A	A	A	A	A	A	A	A
179	T	T	T	T	T	T	T	T	T	T	T	T
180	G	G	G	G	G	G	G	G	G	G	G	G
181	A	A	A	A	A	A	A	A	A	A	A	A
182	C	C	C	C	C	C	C	C	C	C	C	C
183	C	C	C	C	C	C	C	C	C	C	C	C
184	A	A	A	A	A	A	A	A	A	A	A	A
185	A	A	A	A	A	A	A	A	A	A	A	A
186	A	A	A	A	A	A	A	A	A	A	A	A
187	A	A	A	A	A	A	A	A	A	A	A	A
188	T	T	T	T	T	T	T	T	T	T	T	T
189	G	G	G	G	G	G	G	G	G	G	G	G
190	A	A	A	A	A	A	A	A	A	A	A	A
191	G	G	G	G	G	G	G	G	G	G	G	G
192	A	A	A	A	A	A	A	A	A	A	A	A
193	A	A	A	A	A	A	A	A	A	A	A	A
194	G	G	G	G	G	G	G	G	G	G	G	G
195	C	C	C	C	C	C	C	C	C	C	C	C
196	A	A	A	A	A	A	A	A	A	A	A	A
197	C	C	C	C	C	C	C	C	C	C	C	C
198	C	C	C	C	C	C	C	C	C	C	C	C
199	G	G	G	G	G	G	G	G	G	G	G	G
200	T	T	T	T	T	T	T	T	T	T	T	T
201	C	C	C	C	C	C	C	C	C	C	C	C
202	C	C	C	C	C	C	C	C	C	C	C	C
203	T	T	T	T	T	T	T	T	T	T	T	T
204	G	G	G	G	G	G	G	G	G	G	G	G
205	A	A	A	A	A	A	A	A	A	A	A	A
206	T	T	T	T	T	T	T	T	T	T	T	T
207	C	C	C	C	C	C	C	C	C	C	C	C
208	G	G	G	G	G	G	G	G	G	G	G	G
209	T	T	T	T	T	T	T	T	T	T	T	T
210	C	C	C	C	C	C	C	C	C	C	C	C
211	T	T	T	T	T	T	T	T	T	T	T	T
212	G	G	G	G	G	G	G	G	G	G	G	G
213	A	A	A	A	A	A	A	A	A	A	A	A
214	A	A	A	A	A	A	A	A	A	A	A	A
215	A	A	A	A	A	A	A	A	A	A	A	A
216	G	G	G	G	G	G	G	G	G	G	G	G
217	A	A	A	A	A	A	A	A	A	A	A	A
218	A	A	A	A	A	A	A	A	A	A	A	A
219	A	A	A	A	A	A	A	A	A	A	A	A
220	A	A	A	A	A	A	A	A	A	A	A	A
221	C	C	C	C	C	C	C	C	C	C	C	C
222	T	T	T	T	T	T	T	T	T	T	T	T
223	G	G	G	G	G	G	G	G	G	G	G	G
224	A	A	A	A	A	A	A	A	A	A	A	A
225	G	G	G	G	G	G	G	G	G	G	G	G
226	T	T	T	T	T	T	T	T	T	T	T	T
227	C	C	C	C	C	C	C	C	C	C	C	C
228	G	G	G	G	G	G	G	G	G	G	G	G
229	A	A	A	A	A	A	A	A	A	A	A	A
230	C	C	C	C	C	C	C	C	C	C	C	C
231	A	A	A	A	A	A	A	A	A	A	A	A
232	A	A	A	A	A	A	A	A	A	A	A	A
233	C	C	C	C	C	C	C	C	C	C	C	C
234	G	G	G	G	G	G	G	G	G	G	G	G
235	C	C	C	C	C	C	C	C	C	C	C	C

Fig. 7.2 cont.

Fig. 7.2 cont.

DNA SEQUENCING

Position	Tm12.84-2.2	Tm12.84-2.3	Tm12.84-3.4	Tm12.84-3.9	Tm12.84-7.5	Consensus of	Tm13.17	Consensus with	B1	Consensus with	AFP-3	Consensus with
354	A	A	A	A	A	A	R	R	A	N	N	N
355	C	C	C	C	C	C	C	C	C	C/G	C/T	C/G
356	G	G	G	G	G	G	G	G	G	G/C	T/C	G/C
357	G	G	G	G	G	G	G	G	G	N	T/C	N
358	G	G	G	G	G	G	G	G	G	N	A/G	N
359	C	C	C	C	C	C	G	G	G	N	G/C	N
360	T	T	T	T	T	T	T	T	T	N	T/C	N
361	T	T	T	T	T	T	A	A	A	N	A/G	N
362	A	A	A	A	A	A	T	T	T	T	C/C	N
363	T	T	T	T	T	T	A	A	A	R	G/A	R
364	G	G	G	G	G	G	T	T	T	G	C/G	R
365	A	A	A	A	A	A	C	C	C	A	C/G	A
366	A	A	A	A	A	A	G	G	G	N	N	N
367	C	C	C	C	C	C	T	T	T	N	N	N
368	C	C	C	C	C	C	G	G	G	N	N	N
369	C	C	C	C	C	C	T	T	T	N	N	N
370	T	T	T	T	T	T	C	C	C	N	N	N
371	T	T	T	T	T	T	A	A	A	N	N	N
372	C	C	C	C	C	C	T	T	T	N	N	N
373	A	A	A	A	A	A	C	C	C	N	N	N
374	A	A	A	A	A	A	T	T	T	N	N	N
375	G	G	G	G	G	G	G	G	G	N	N	N
376	T	T	T	T	T	T	T	T	T	N	N	N
377	G	G	G	G	G	G	T	T	T	N	N	N
378	T	T	T	T	T	T	A	A	A	N	N	N
379	T	T	T	T	T	T	T	T	T	N	N	N
380	T	T	T	T	T	T	A	A	A	N	N	N
381	T	T	T	T	T	T	T	T	T	N	N	N
382	T	T	T	T	T	T	A	A	A	N	N	N
383	A	A	A	A	A	A	C	C	C	N	N	N
384	C	C	C	C	C	C	G	G	G	N	N	N
385	G	G	G	G	G	G	A	A	A	N	N	N
386	A	A	A	A	A	A	C	C	C	N	N	N
387	C	C	C	C	C	C	G	G	G	N	N	N
388	A	A	A	A	A	A	A	A	A	N	N	N
389	G	G	G	G	G	G	A	A	A	N	N	N
390	C	C	C	C	C	C	C	C	C	N	N	N
391	A	A	A	A	A	A	A	A	A	N	N	N
392	A	A	A	A	A	A	A	A	A	N	N	N
393	A	A	A	A	A	A	A	A	A	N	N	N
394	C	C	C	C	C	C	C	C	C	N	N	N
395	C	C	C	C	C	C	C	C	C	N	N	N
396	T	T	T	T	T	T	T	T	T	N	N	N
397	G	G	G	G	G	G	G	G	G	N	N	N
398	A	A	A	A	A	A	G	G	G	N	N	N
399	T	T	T	T	T	T	T	T	T	N	N	N
400	T	T	T	T	T	T	T	T	T	N	N	N
401	T	T	T	T	T	T	T	T	T	N	N	N
402	C	C	C	C	C	C	T	T	T	N	N	N
403	T	T	T	T	T	T	C	C	C	N	N	N
404	T	T	T	T	T	T	C	C	C	N	N	N
405	C	C	C	C	C	C	G	G	G	N	N	N
406	T	T	T	T	T	T	A	A	A	N	N	N
407	T	T	T	T	T	T	T	T	T	N	N	N
408	G	G	G	G	G	G	G	G	G	N	N	N
409	C	C	C	C	C	C	C	C	C	N	N	N
410	A	A	A	A	A	A	A	A	A	N	N	N
411	T	T	T	T	T	T	T	T	T	N	N	N
412	T	T	T	T	T	T	T	T	T	N	N	N
413	G	G	G	G	G	G	G	G	G	N	N	N
414	A	A	A	A	A	A	A	A	A	N	N	N
415	T	T	T	T	T	T	T	T	T	N	N	N
416	T	T	T	T	T	T	T	T	T	N	N	N
417	T	T	T	T	T	T	T	T	T	N	N	N
418	A	A	A	A	A	A	A	A	A	N	N	N
419	A	A	A	A	A	A	A	A	A	N	N	N
420	T	T	T	T	T	T	T	T	T	N	N	N
421	G	G	G	G	G	G	G	G	G	N	N	N
422	T	T	T	T	T	T	T	T	T	N	N	N
423	T	T	T	T	T	T	T	T	T	N	N	N
424	G	G	G	G	G	G	G	G	G	N	N	N
425	T	T	T	T	T	T	T	T	T	N	N	N
426	T	T	T	T	T	T	T	T	T	N	N	N
427	G	G	G	G	G	G	G	G	G	N	N	N
428	T	T	T	T	T	T	T	T	T	N	N	N
429	T	T	T	T	T	T	T	T	T	N	N	N
430	T	T	T	T	T	T	T	T	T	N	N	N
431	T	T	T	T	T	T	T	T	T	N	N	N
432	G	G	G	G	G	G	G	G	G	N	N	N
433	A	A	A	A	A	A	A	A	A	N	N	N
434	C	C	C	C	C	C	C	C	C	N	N	N
435	G	G	G	G	G	G	G	G	G	N	N	N
436	T	T	T	T	T	T	T	T	T	N	N	N
437	G	G	G	G	G	G	G	G	G	N	N	N
438	A	A	A	A	A	A	A	A	A	N	N	N
439	A	A	A	A	A	A	A	A	A	N	N	N
440	T	T	T	T	T	T	T	T	T	N	N	N
441	T	T	T	T	T	T	T	T	T	N	N	N
442	T	T	T	T	T	T	T	T	T	N	N	N
443	G	G	G	G	G	G	G	G	G	N	N	N
444	A	A	A	A	A	A	A	A	A	N	N	N
445	G	G	G	G	G	G	G	G	G	N	N	N
446	A	A	A	A	A	A	A	A	A	N	N	N
447	T	T	T	T	T	T	T	T	T	N	N	N
448	T	T	T	T	T	T	T	T	T	N	N	N
449	T	T	T	T	T	T	T	T	T	N	N	N
450	T	T	T	T	T	T	T	T	T	N	N	N
451	T	T	T	T	T	T	T	T	T	N	N	N
452	G	G	G	G	G	G	G	G	G	N	N	N
453	T	T	T	T	T	T	T	T	T	N	N	N
454	T	T	T	T	T	T	T	T	T	N	N	N
455	T	T	T	T	T	T	T	T	T	N	N	N
456	T	T	T	T	T	T	T	T	T	N	N	N
457	T	T	T	T	T	T	T	T	T	N	N	N
458	G	G	G	G	G	G	G	G	G	N	N	N
459	A	A	A	A	A	A	A	A	A	N	N	N
460	G	G	G	G	G	G	G	G	G	N	N	N
461	T	T	T	T	T	T	T	T	T	N	N	N
462	T	T	T	T	T	T	T	T	T	N	N	N
463	T	T	T	T	T	T	T	T	T	N	N	N
464	T	T	T	T	T	T	T	T	T	N	N	N
465	T	T	T	T	T	T	T	T	T	N	N	N
466	T	T	T	T	T	T	T	T	T	N	N	N
467	T	T	T	T	T	T	T	T	T	N	N	N
468	T	T	T	T	T	T	T	T	T	N	N	N
469	T	T	T	T	T	T	T	T	T	N	N	N
470	G	G	G	G	G	G	G	G	G	N	N	N
471	A	A	A	A	A	A	A	A	A	N	N	N

Fig. 7.2 cont.

Position	Tm12.84-2.2	Tm12.84-2.3	Tm12.84-3.4	Tm12.84-3.9	Tm12.84-7.5	Consensus of	Tm13.17	Consensus with	B1	Consensus with	AFP-3	Consensus with
472	A	A	A	A	A	A	A	A	A	A	A	A
473	A	A	A	A	A	A	A	A	A	A	A	A
474	T	T	T	T	T	T	T	T	T	T	T	T
475	A	A	A	A	A	A	A	A	A	A	A	A
476	A	A	A	A	A	A	A	A	A	A	A	A
477	A	A	A	A	A	A	A	A	A	A	A	A
478	G	G	G	G	G	G	G	G	N	N	T	N
479	T	T	T	T	T	T	T	N	N	N	T	N
480	A	A	C	C	C	C	C	Y	Y	A	A	N
481	A	C	C	C	T	T	T	T	T	A	A	N
482	A	T	T	T	T	T	T	T	T	T	A	A
483	T	T	T	T	T	T	T	T	T	T	A	AT
484	A	A	A	A	A	A	T	T	T	T	G	AT
485	T	T	T	T	T	T	T	T	T	T	A	AT
486	C	C	C	G	G	G	G	G	R	R	A	R
487	G	G	G	G	G	G	G	G	R	R	A	T/A
488	T	T	T	T	T	T	T	T	T	T	A	N
489	T	T	T	T	T	T	T	T	N	N	A	N
490	A	A	A	A	A	A	A	N	N	N	A	N
491	T	T	G	G	T	G	T	N	N	R	A	N
492	G	G	G	G	G	G	G	N	N	R	A	N
493	T	A	A	A	T	A	A	A	A	A	A	A
494	A	A	A	A	A	A	A	A	A	A	A	A
495	A	A	A	A	A	A	A	A	A	A	A	A
496	A	A	A	A	A	A	A	A	A	A	A	A
497	A	A	A	A	A	A	A	A	A	A	A	A
498	A	A	A	A	A	A	A	A	A	A	A	A
499	A	A	A	A	A	A	A	A	A	A	A	A
500	A	A	A	A	A	A	A	A	A	A	A	A
501	A	A	A	A	A	A	A	A	A	A	A	A
502	A	A	A	A	A	A	A	A	A	A	A	A
503	A	A	A	A	A	A	A	A	A	A	A	A
504	A	A	A	A	A	A	A	A	A	A	A	A
505	A	A	A	A	A	A	A	A	A	A	A	A
506	A	A	A	A	A	A	A	A	A	A	A	A
507	A	A	A	A	A	A	A	A	A	A	A	A
508	A	A	A	A	A	A	A	A	A	A	A	A
509	A	A	A	A	A	A	A	A	A	A	A	A
510	A	A	A	A	A	A	A	A	A	A	A	A
511	A	A	A	A	A	A	A	A	A	A	A	A
512	A	A	A	A	A	A	A	A	A	A	A	A

Fig. 7.2 cont.

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50 51 52 53 54 55 56 57 58 59 60 61 62 63 64 65 66 67 68 69 70 71 72 73 74 75 76 77 78

Position	Tm 1244-2.3	Tm 1244-3.4	Tm 1244-3.9	Tm 1244-7.6	Tm 1244-7.8	Tm 1244-12.4	Tm 13.17	Tm P-81	Tm P-82	Tm AFP-3	GENERAL CONSENSUS	SUBSTITUTIONS - most to least common
1	M	M	M	M	M	M	M	K	K	M	M	K
2	K	K	K	K	K	K	K	L	L	K	K	L
3	L	C	C	L	L	L	L	L	L	L	L	C
4	C	C	C	C	C	C	C	C	C	C	C	C
5	G	G	G	G	G	G	G	G	G	G	G	G
6	H	H	H	H	H	H	H	H	H	H	H	H
7	A	A	A	A	A	A	A	A	A	A	A	A
8	P	P	P	P	P	P	P	P	P	P	P	P
9	R	R	R	R	R	R	R	R	R	R	R	R
10	D	D	D	D	D	D	D	D	D	D	D	D
11	N	N	N	N	N	N	N	N	N	N	N	N
12	T	T	T	T	T	T	T	T	T	T	T	T
13	S	S	S	S	S	S	S	S	S	S	S	S
14	E	E	E	E	E	E	E	E	E	E	E	E
15	T	T	T	T	T	T	T	T	T	T	T	T
16	S	S	S	S	S	S	S	S	S	S	S	S
17	F	F	F	F	F	F	F	F	F	F	F	F
18	I	I	I	I	I	I	I	I	I	I	I	I
19	V	V	V	V	V	V	V	V	V	V	V	V
20	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y	Y
21	W	W	W	W	W	W	W	W	W	W	W	W
22	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
23	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
24	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
25	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
26	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
27	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
28	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
29	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
30	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
31	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
32	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
33	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
34	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
35	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
36	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
37	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
38	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
39	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
40	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
41	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
42	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
43	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
44	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
45	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
46	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
47	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
48	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
49	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
50	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
51	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
52	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
53	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
54	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
55	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
56	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
57	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
58	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
59	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
60	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
61	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
62	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
63	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
64	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
65	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
66	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
67	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
68	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
69	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
70	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
71	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
72	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
73	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
74	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
75	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
76	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
77	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
78	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q

Fig. 7.9

1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49 50 51 52 53 54 55 56 57 58 59 60 61 62 63 64 65 66 67 68 69 70 71 72 73 74 75 76 77 78

Fig. 7.3 cont.

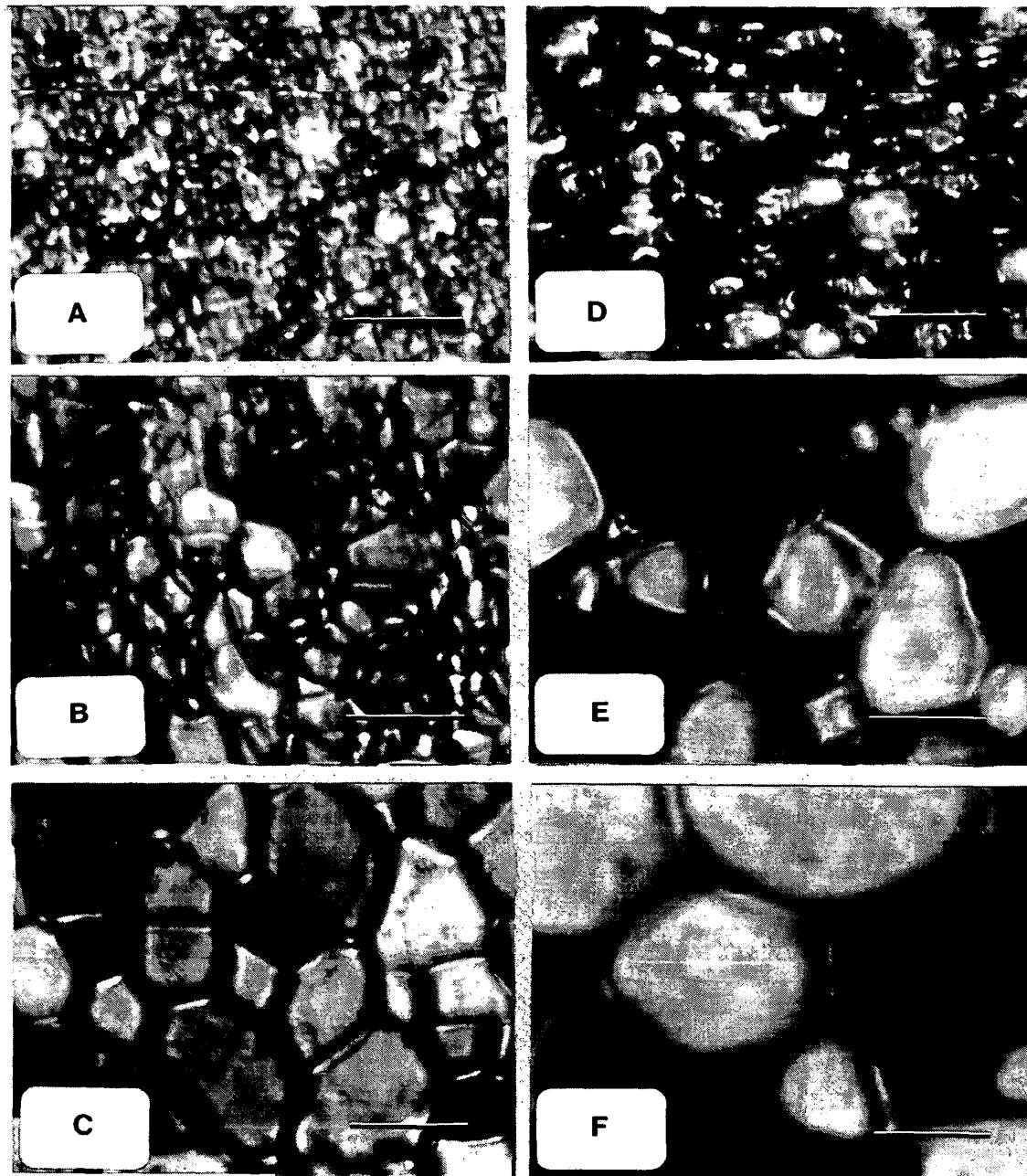


Fig. 8.0

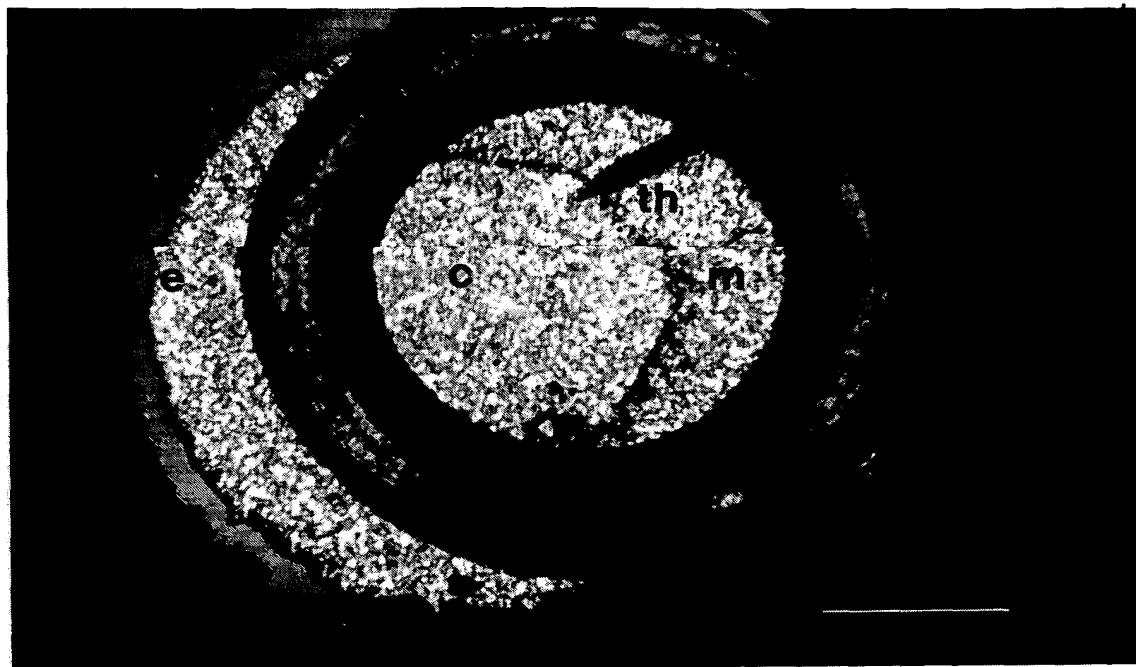


Fig. 8.1a

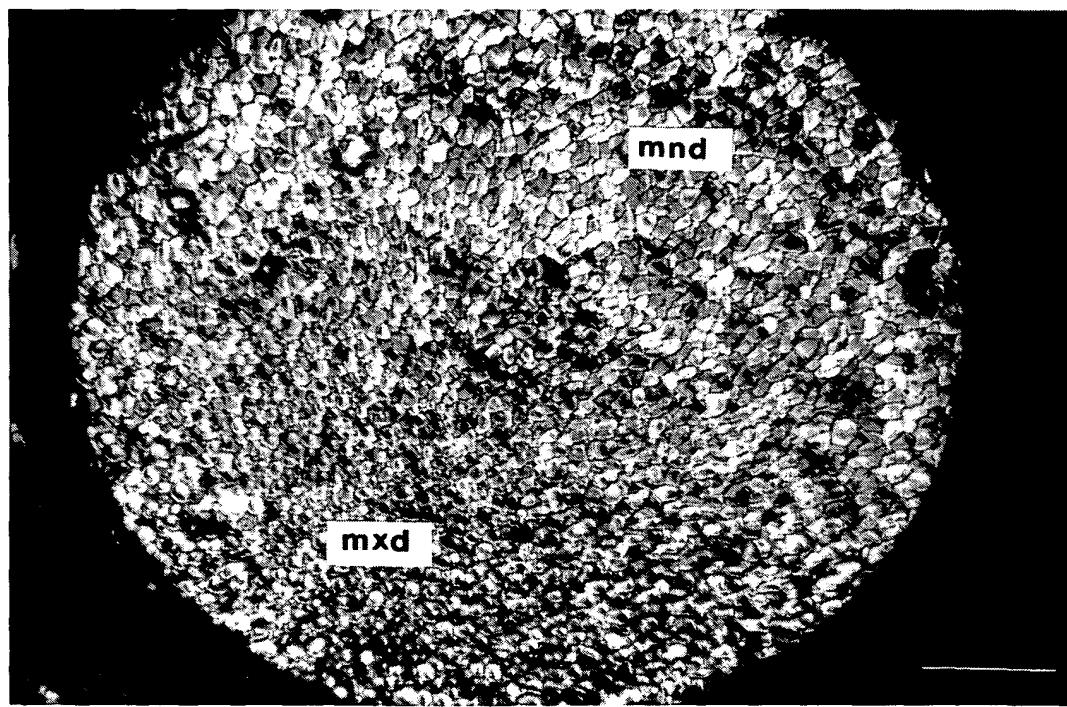
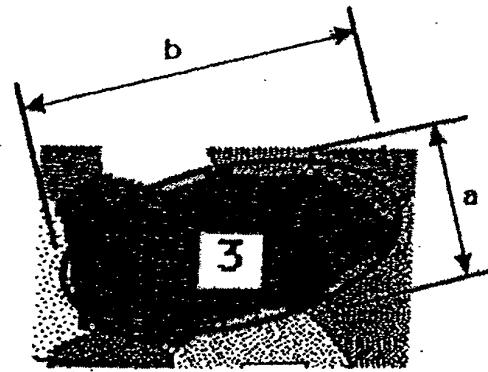
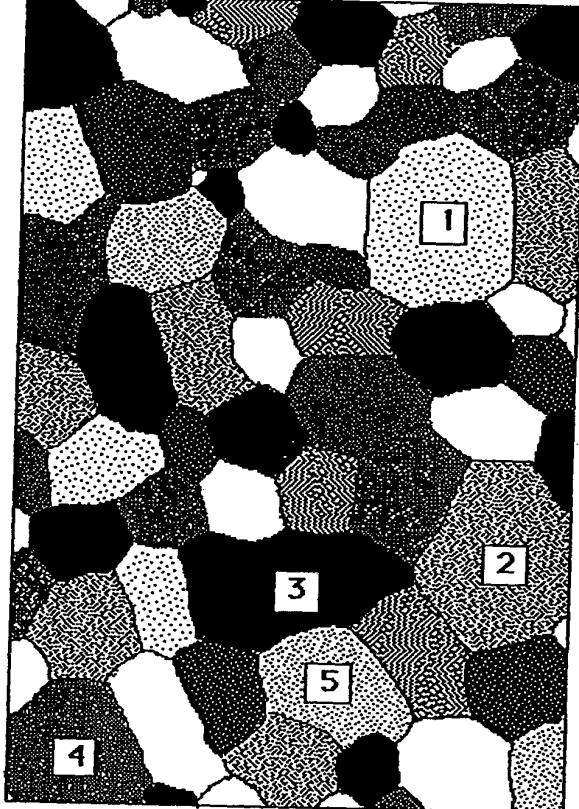


Fig. 8.1b



grain area=0.25 \times a b

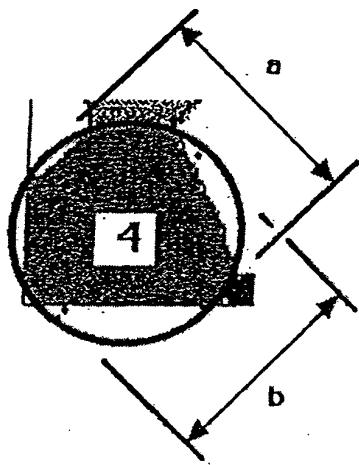


Fig. 8.2

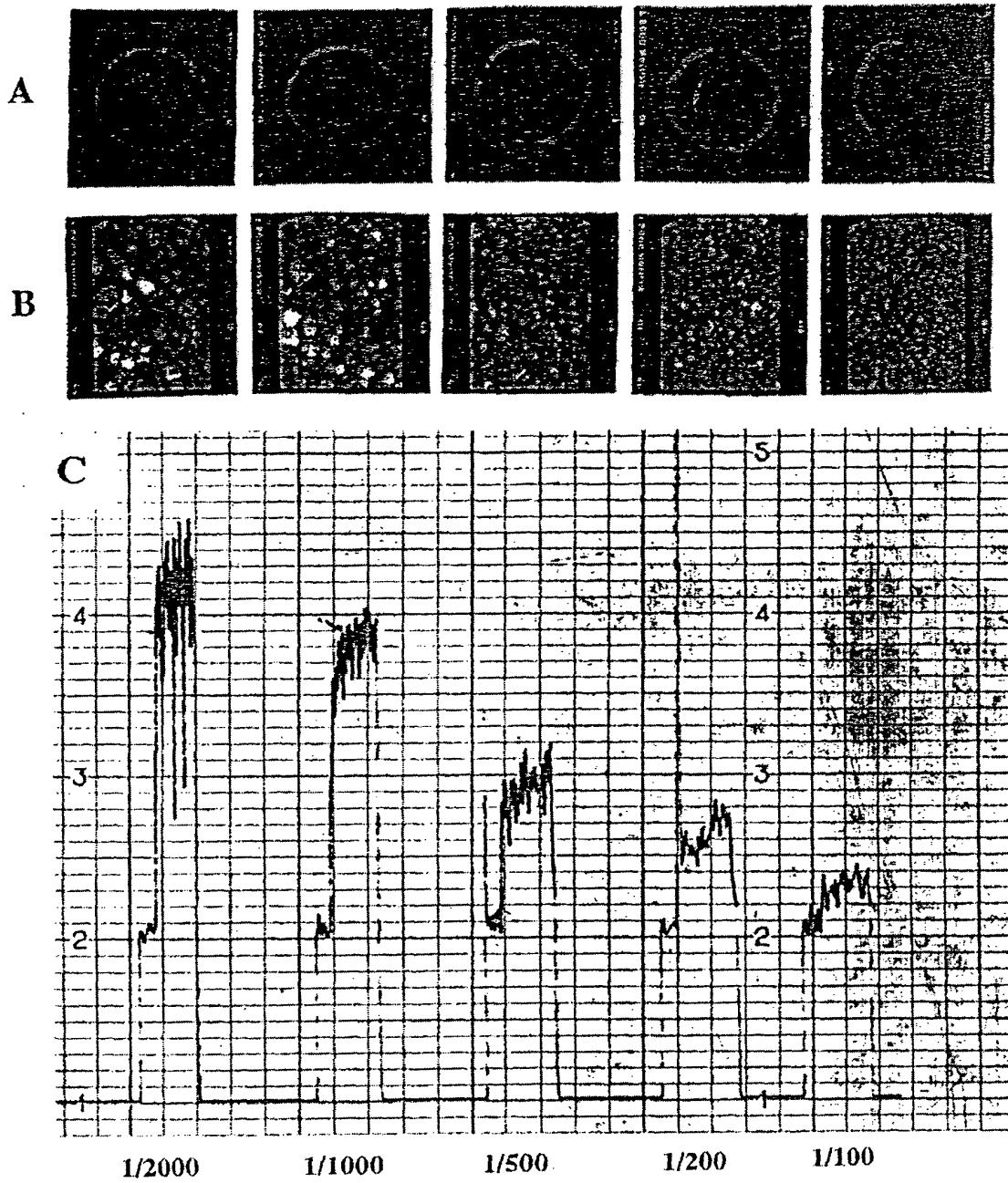


Fig. 8.3

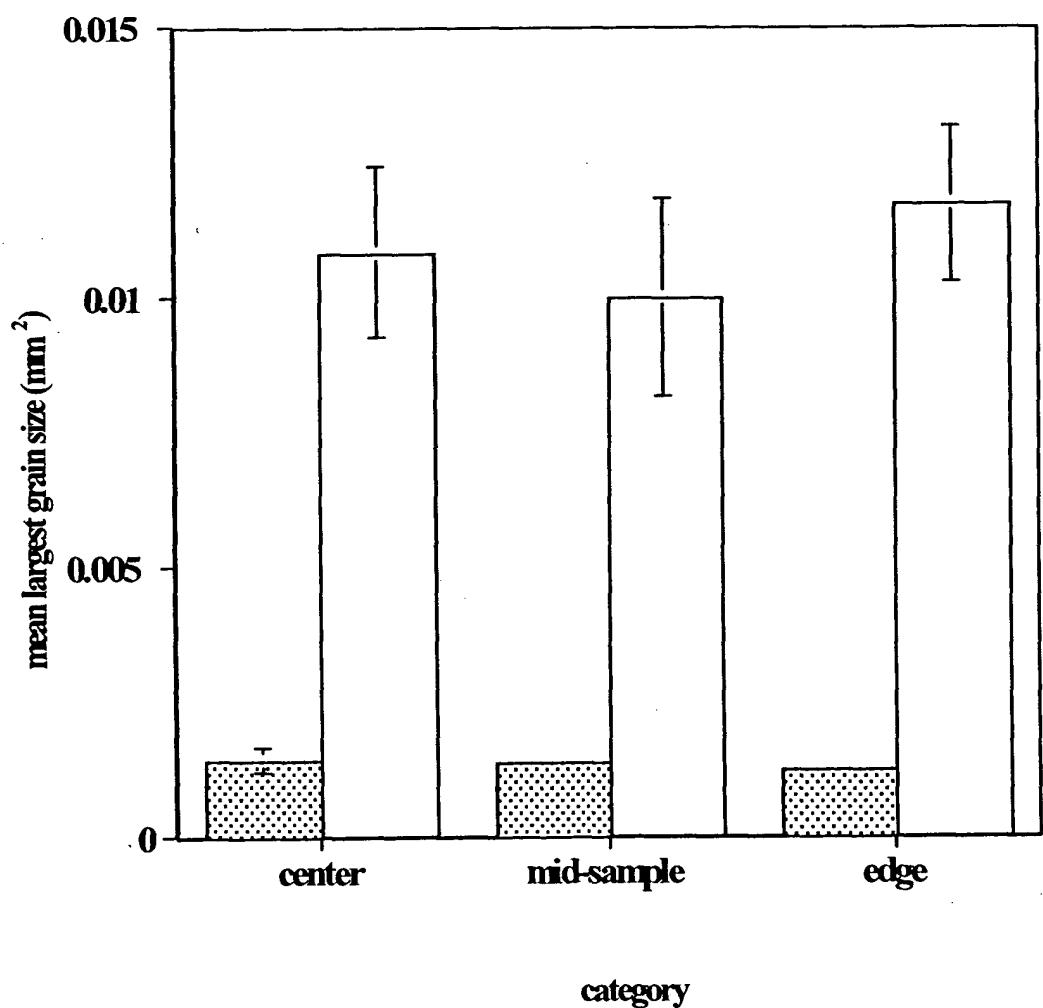


Fig. 8.4a

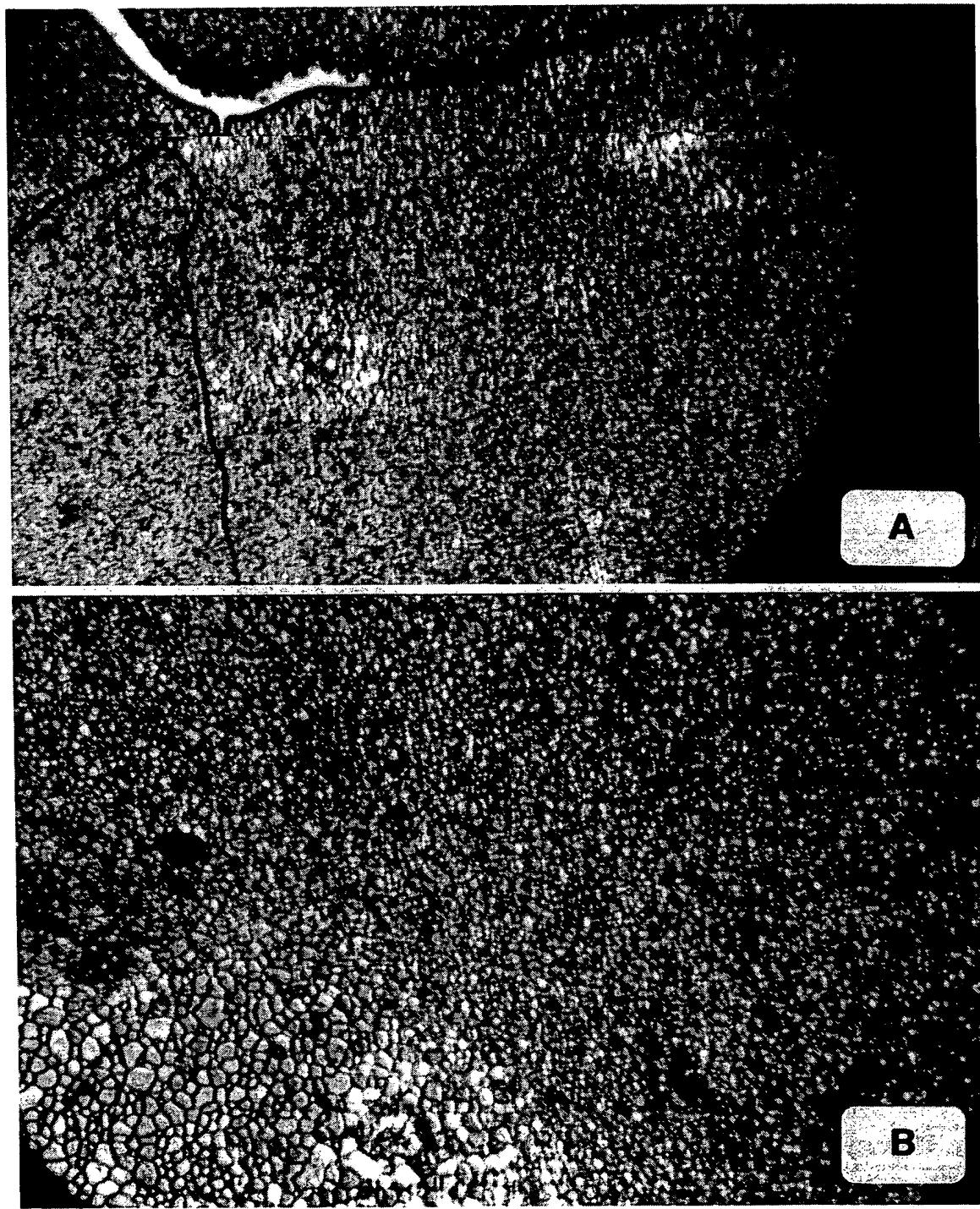


Fig. 8.4b

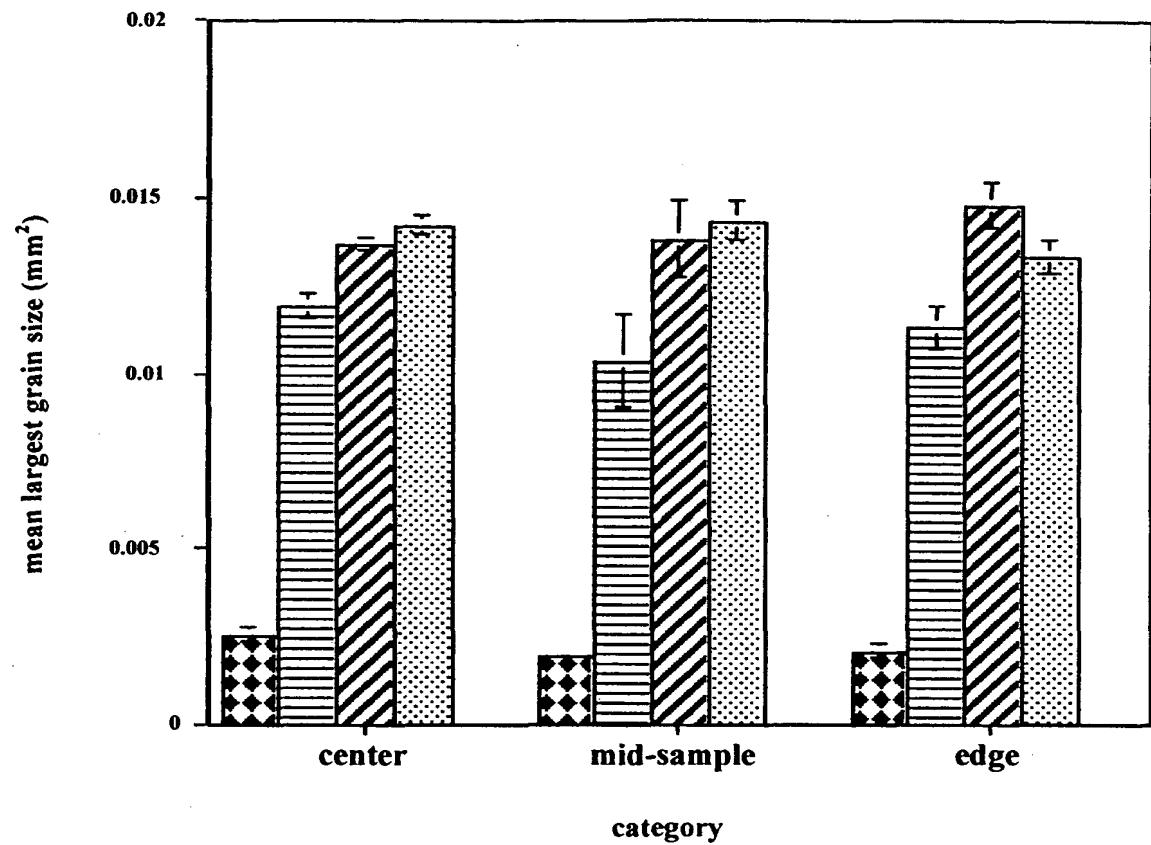


Fig. 8.5a

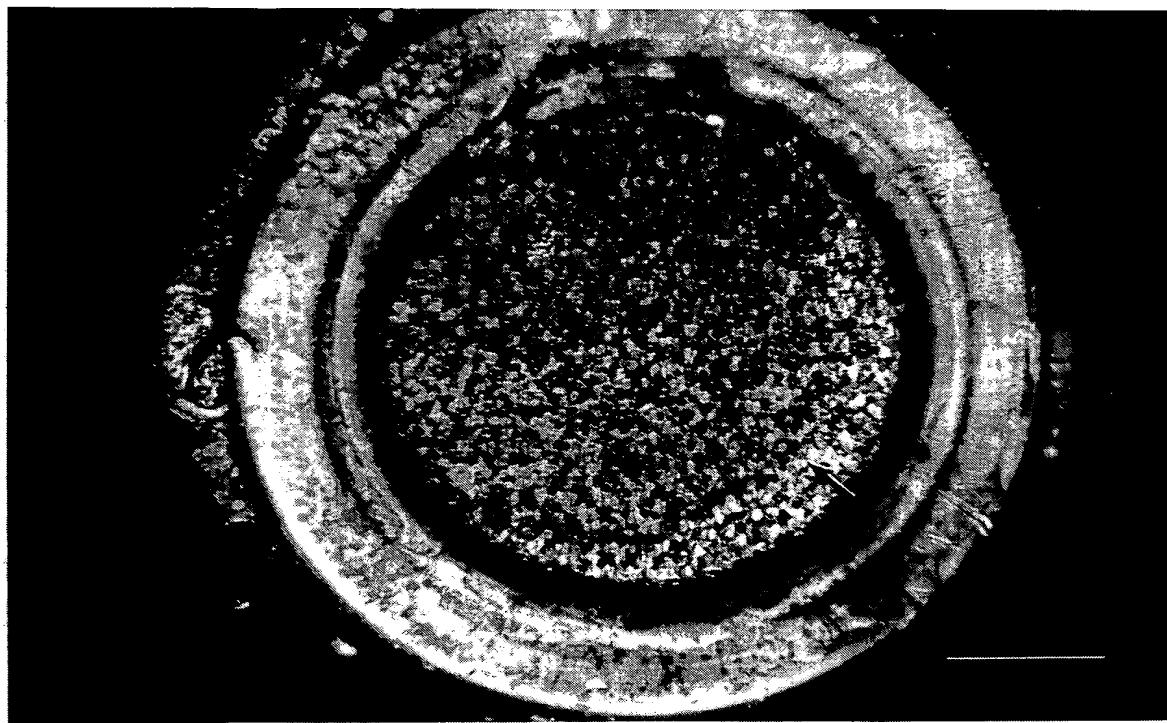


Fig. 8.5b

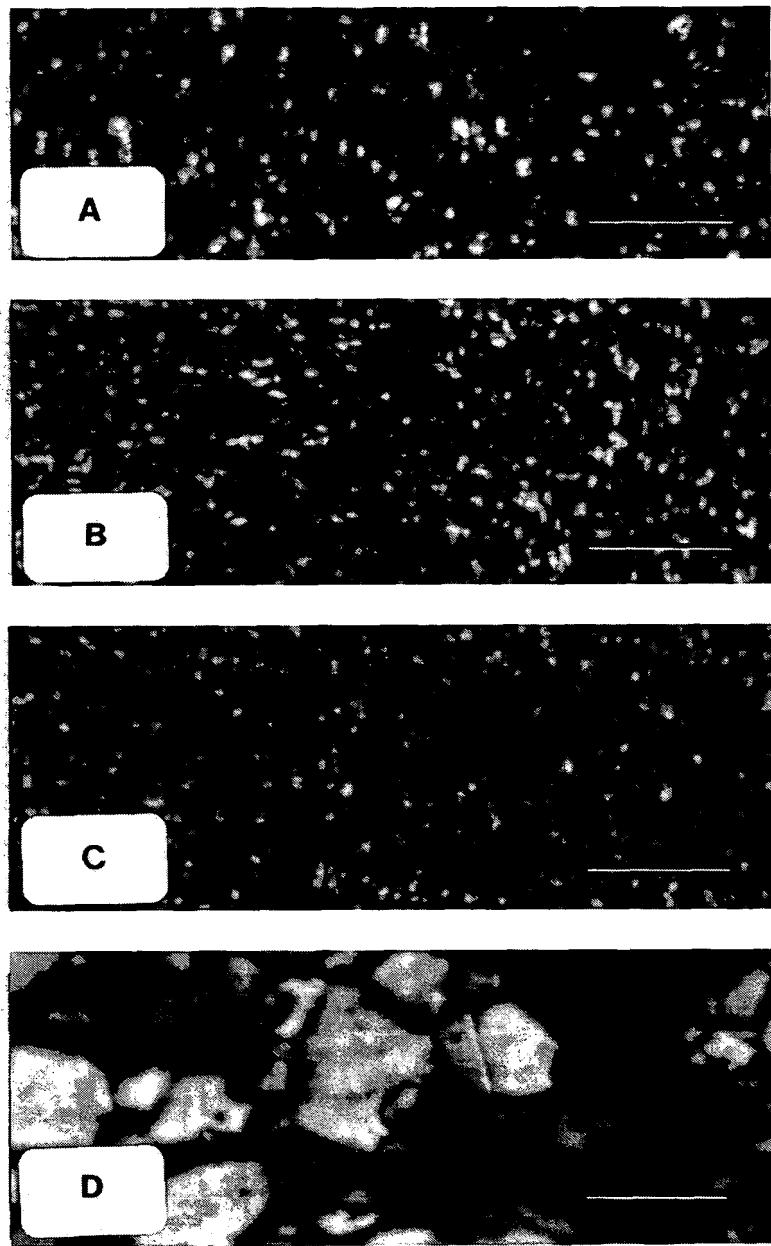


Fig. 8.6

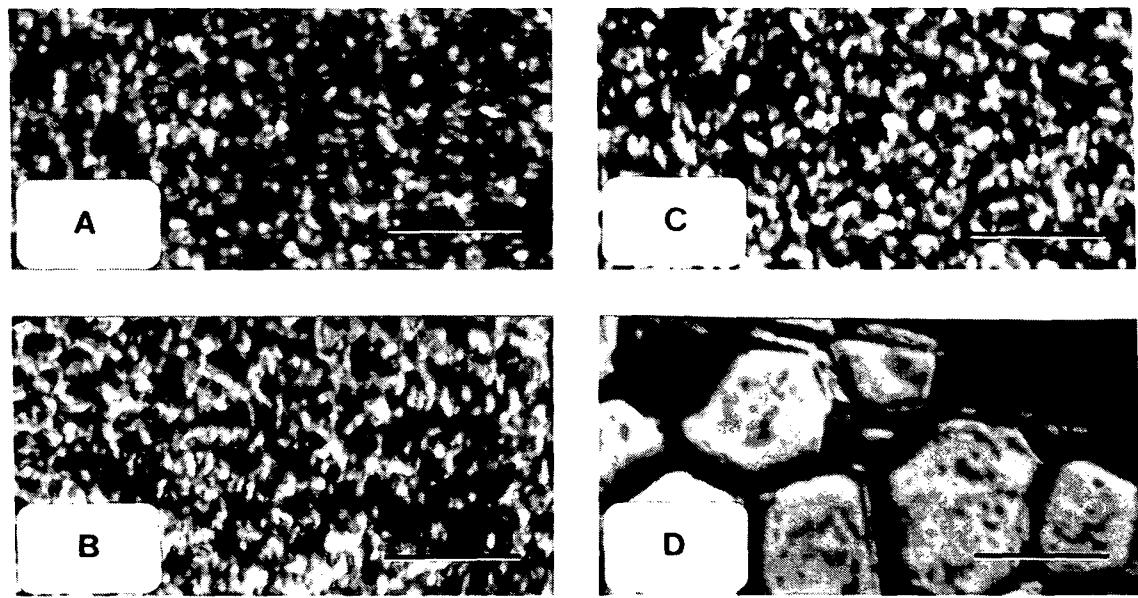


Fig. 8.7

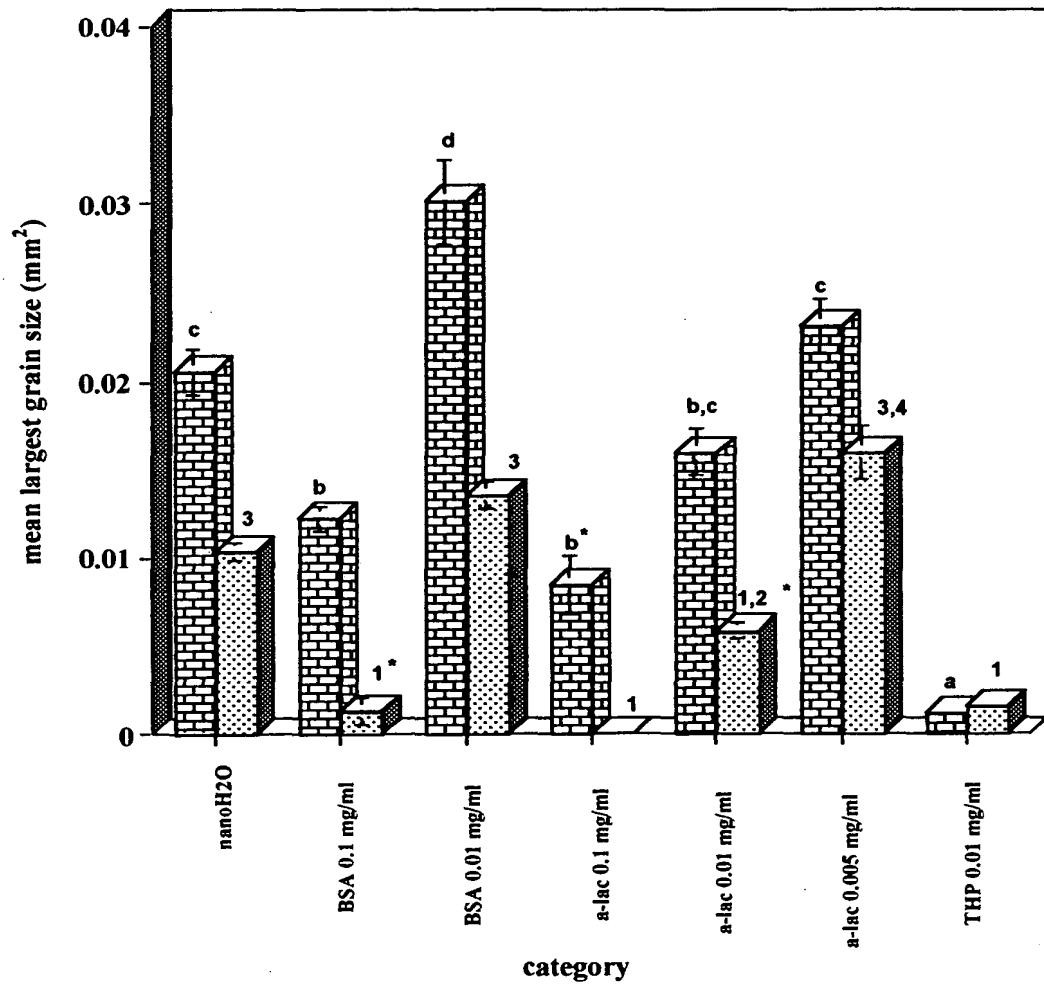


Fig. 8.8

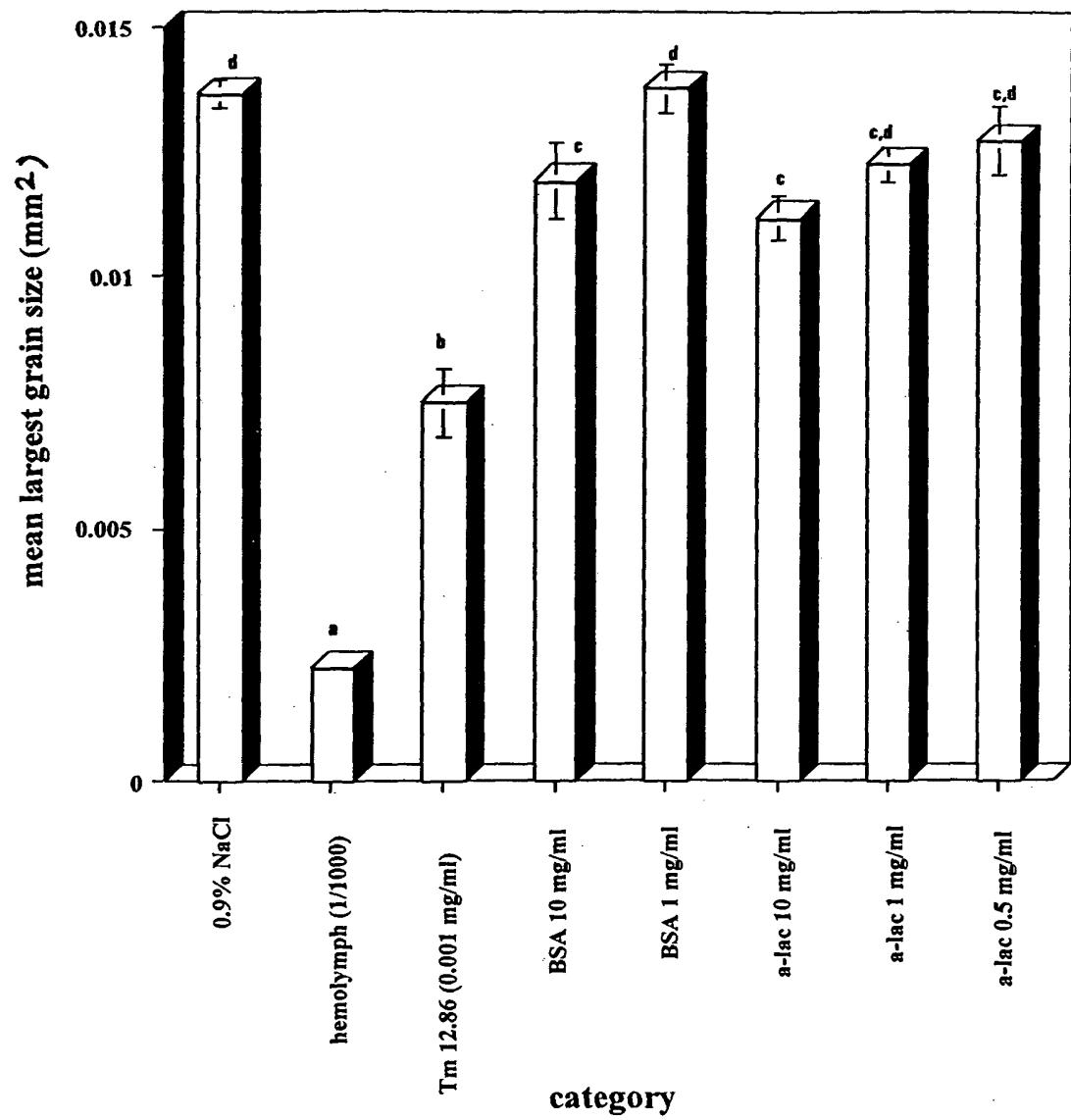


Fig. 8.9

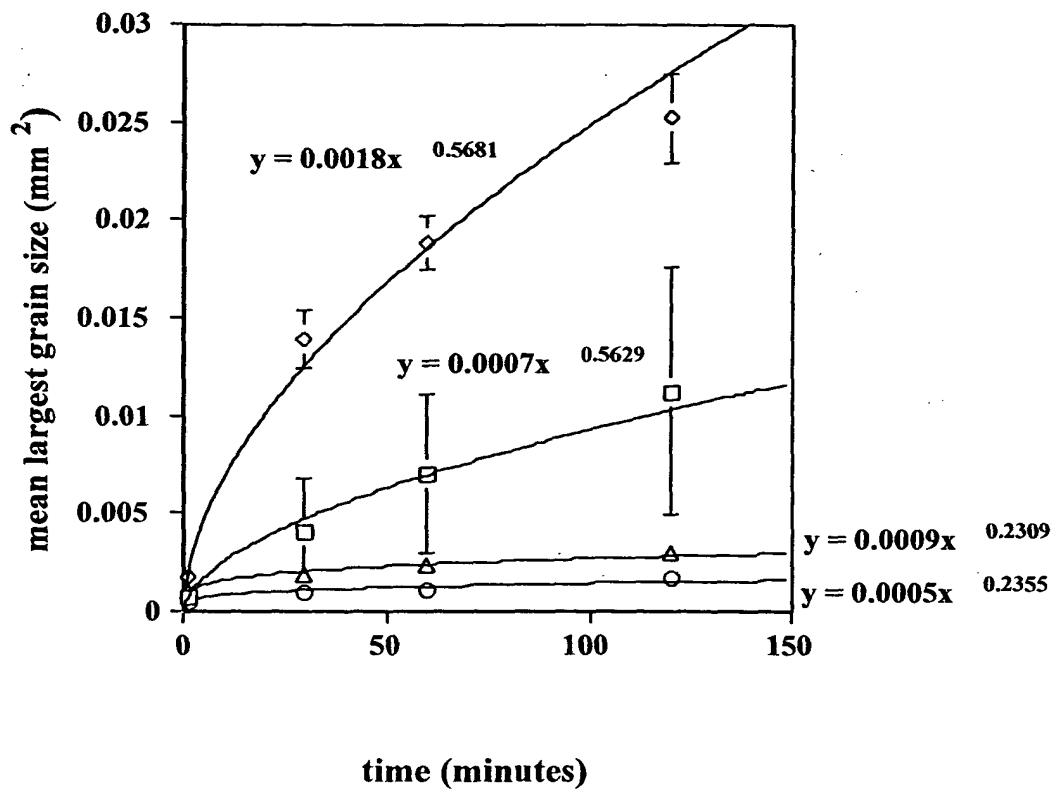


Fig. 8.10

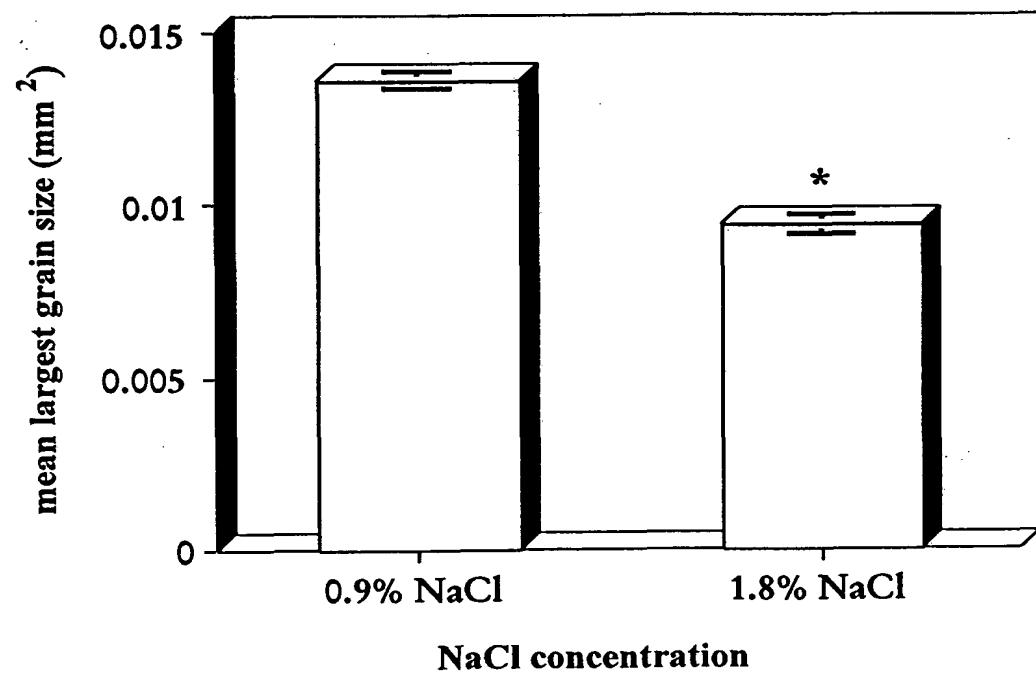


Fig. 8.11

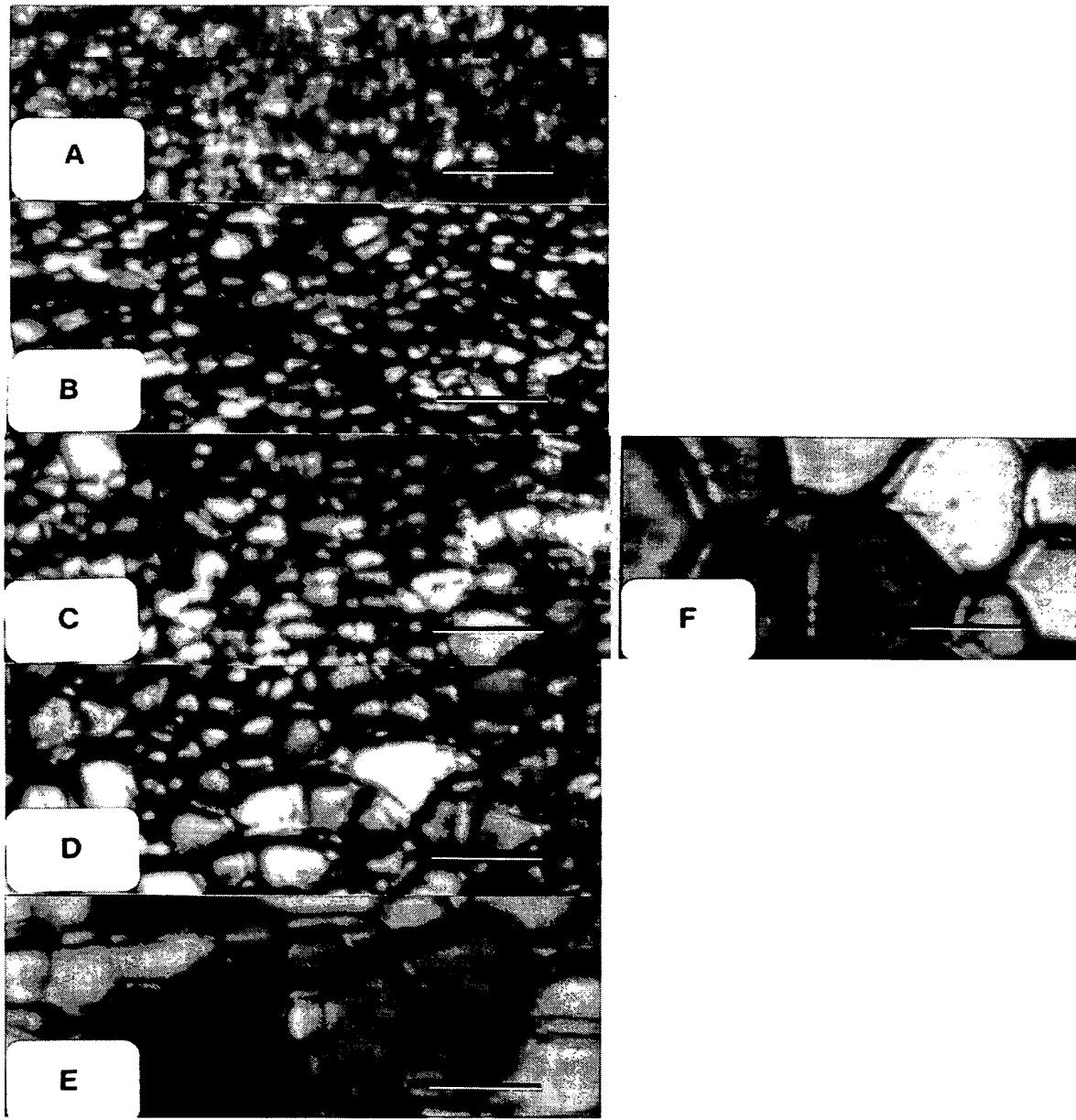


Fig. 8.12

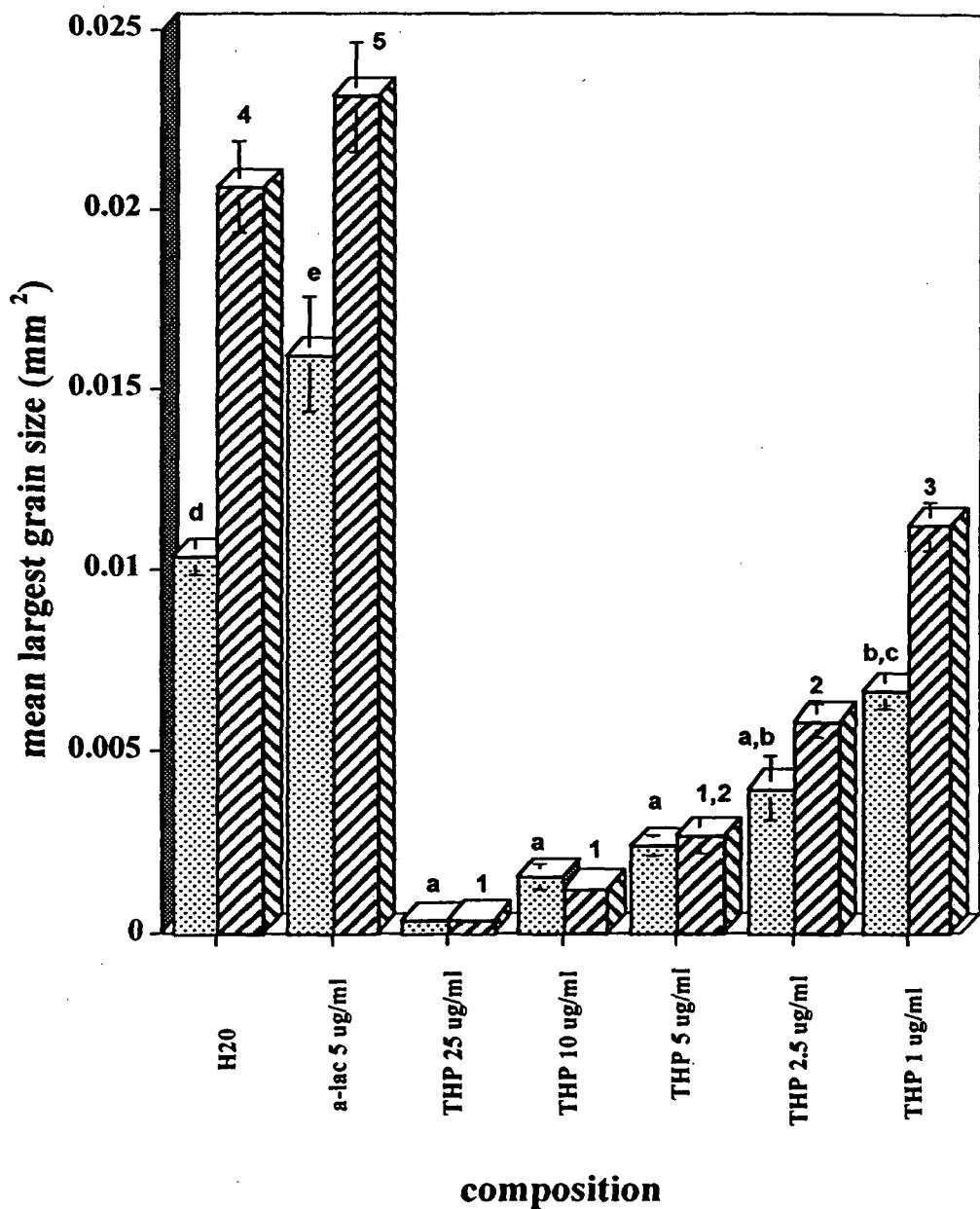


Fig. 8.13

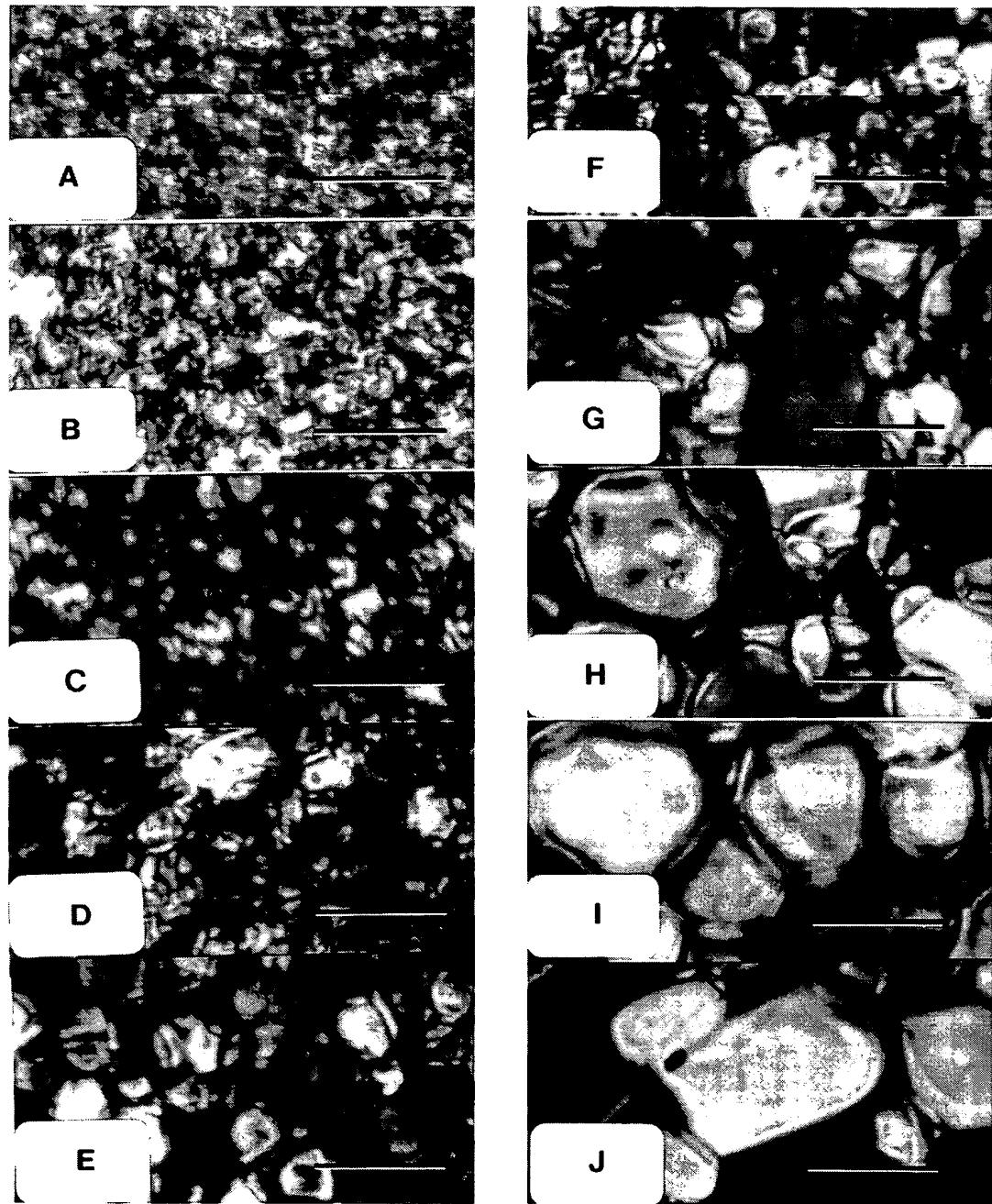


Fig. 8.14

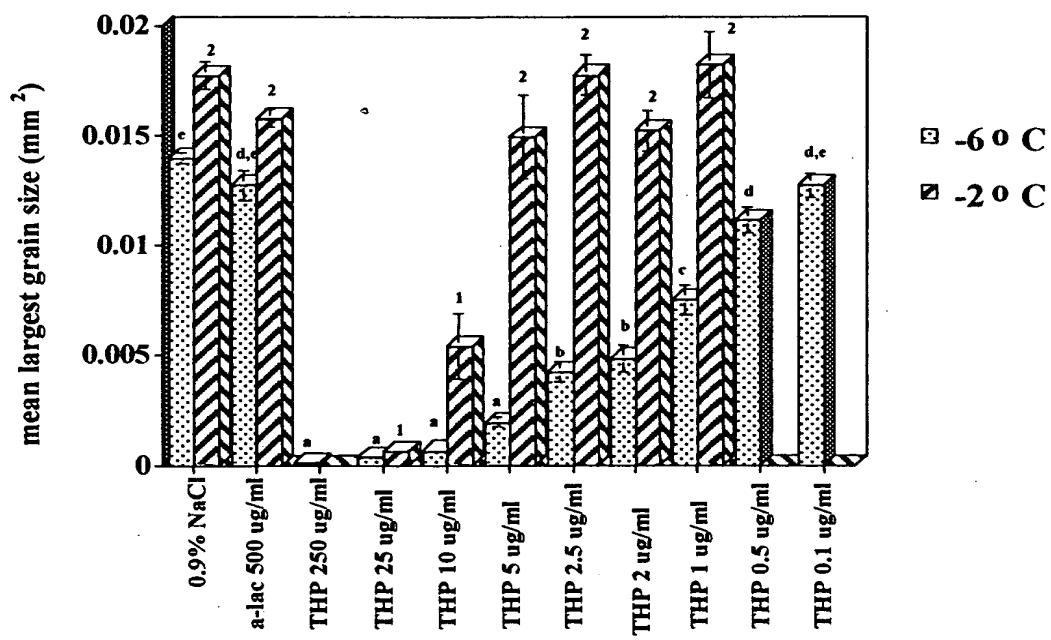


Fig. 8.15

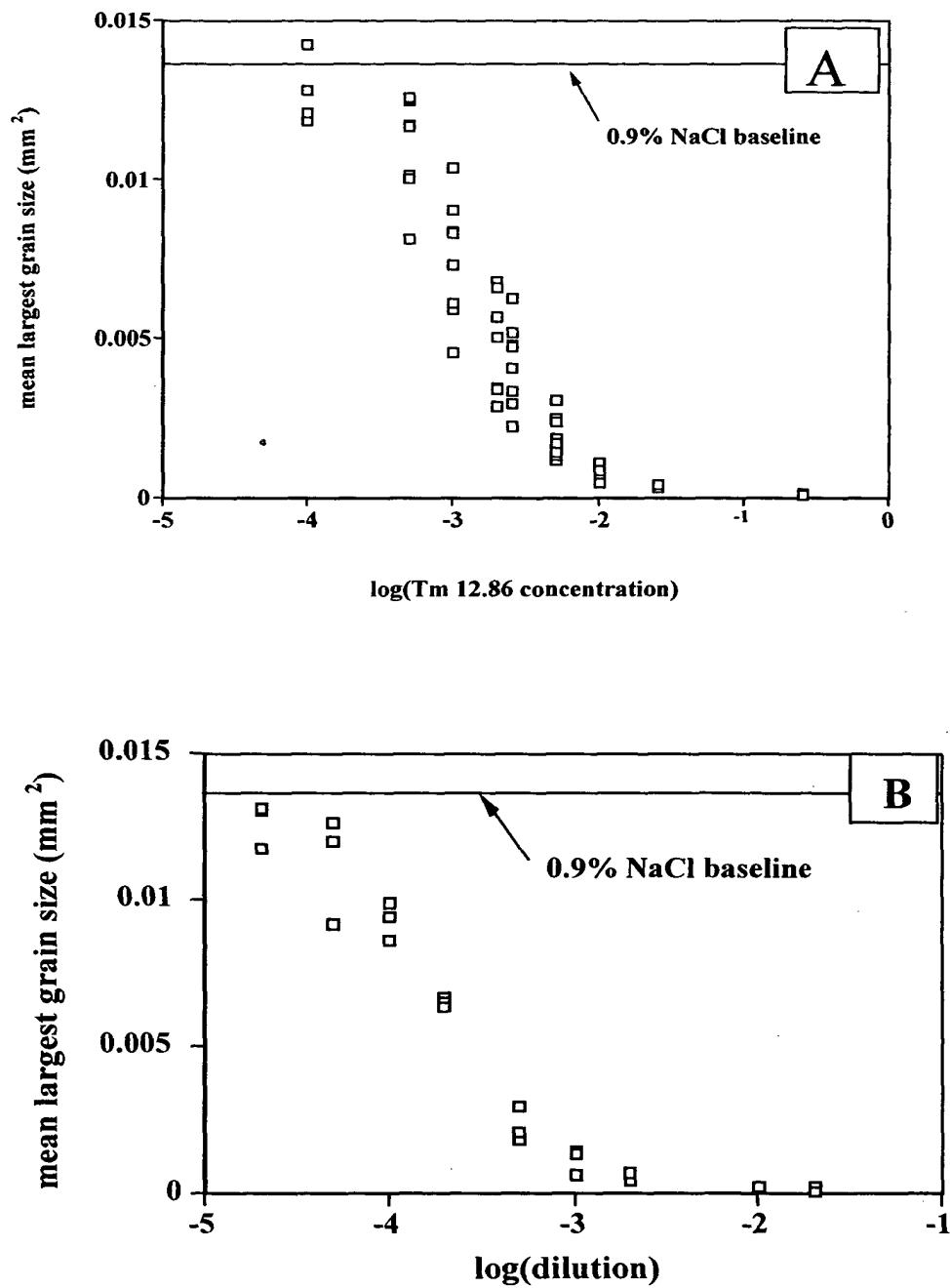


Fig. 8.16

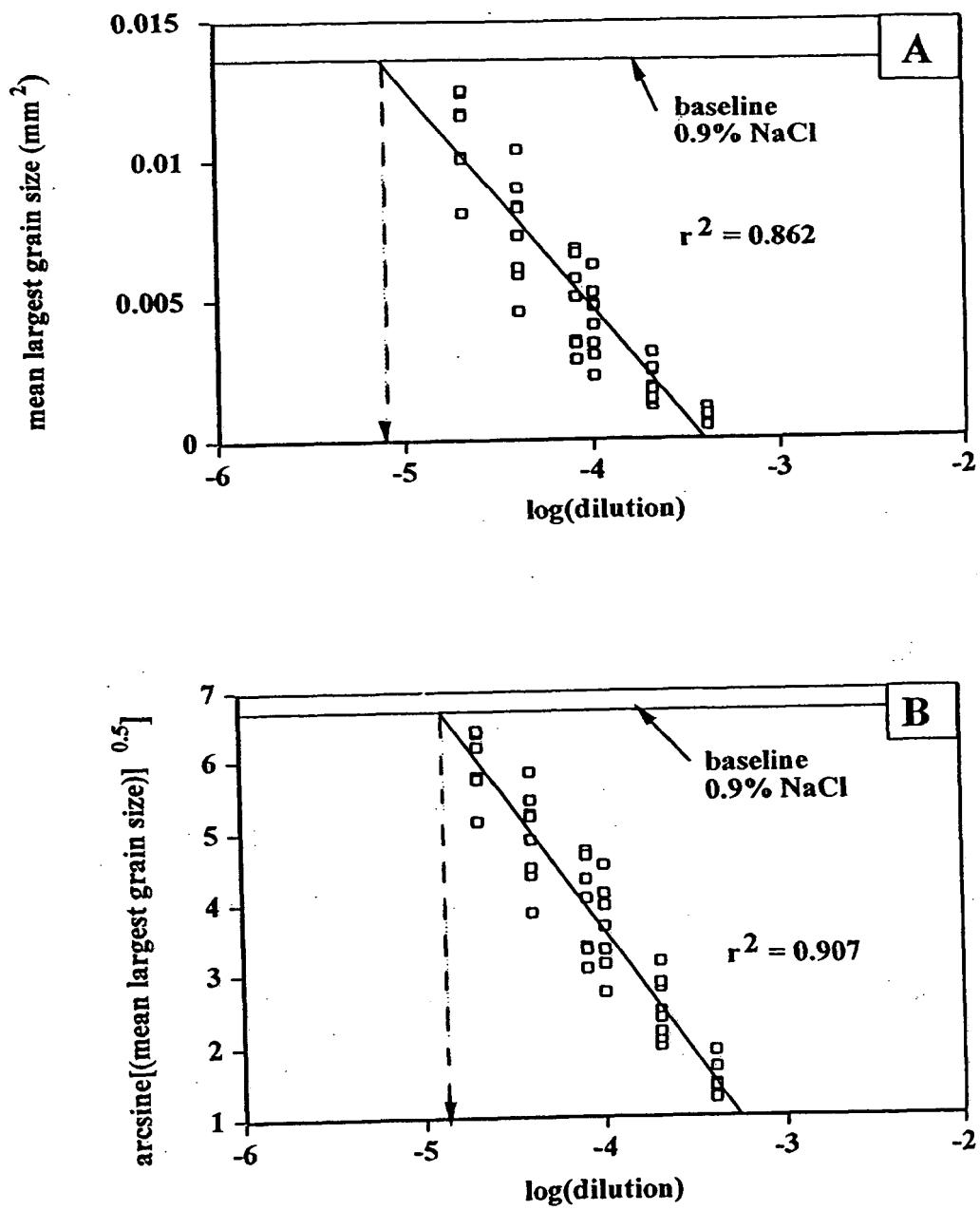


Fig. 8.17

100 90 80 70 60 50 40 30 20 10

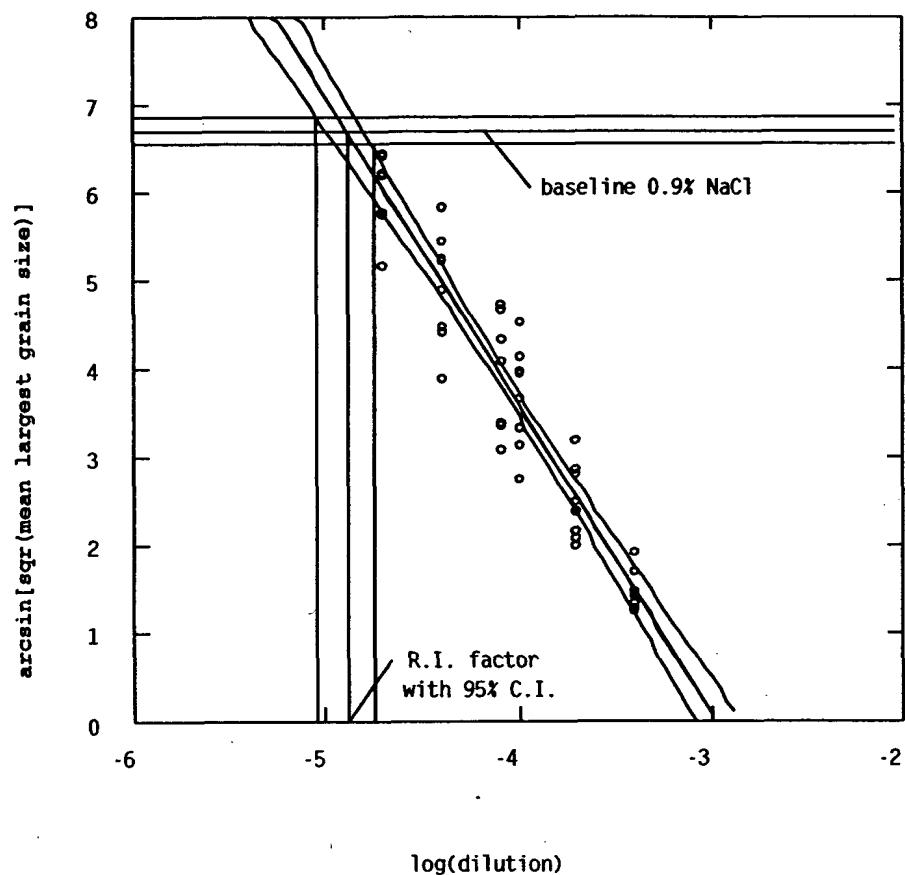


Fig. 8.18

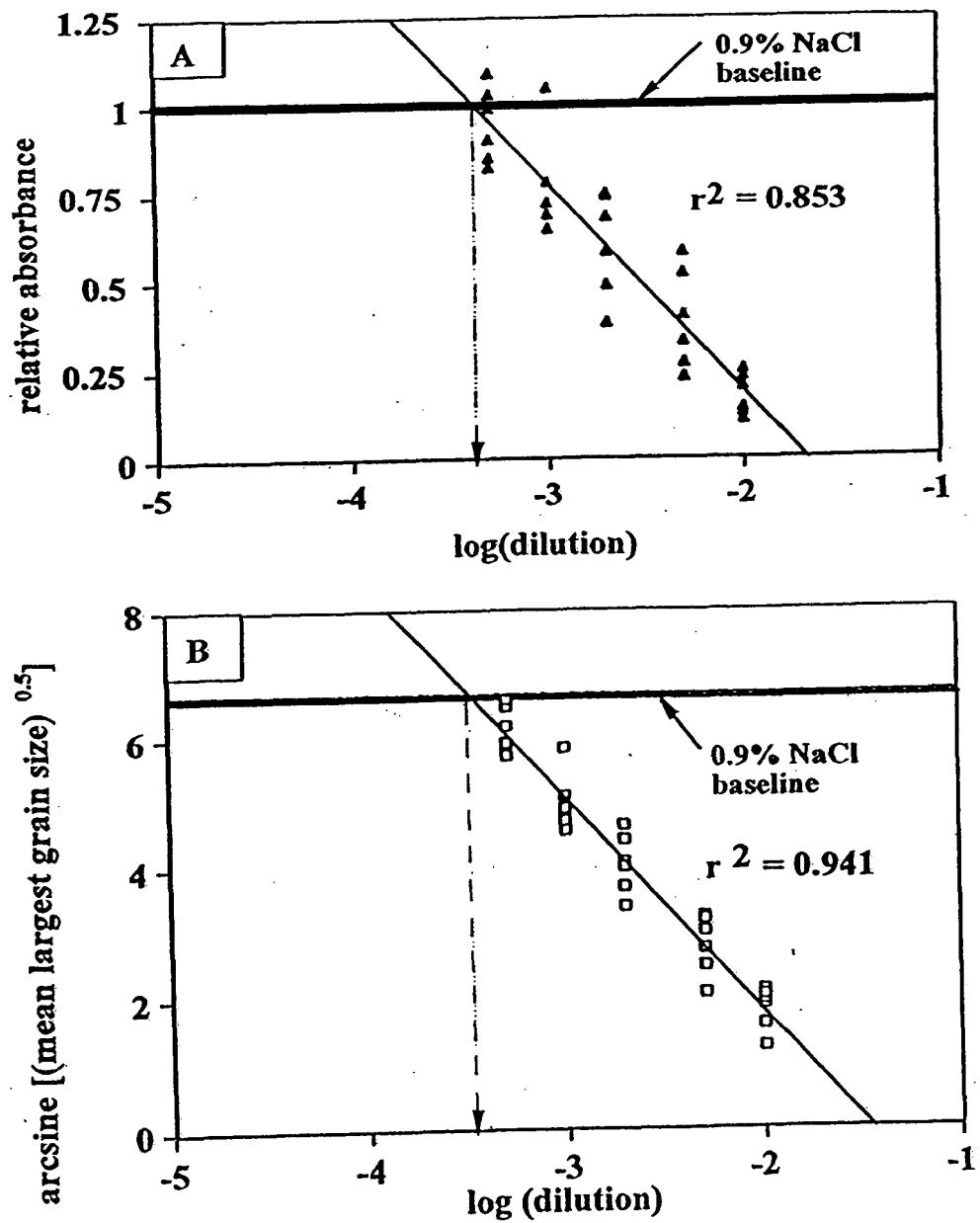


Fig. 8.19

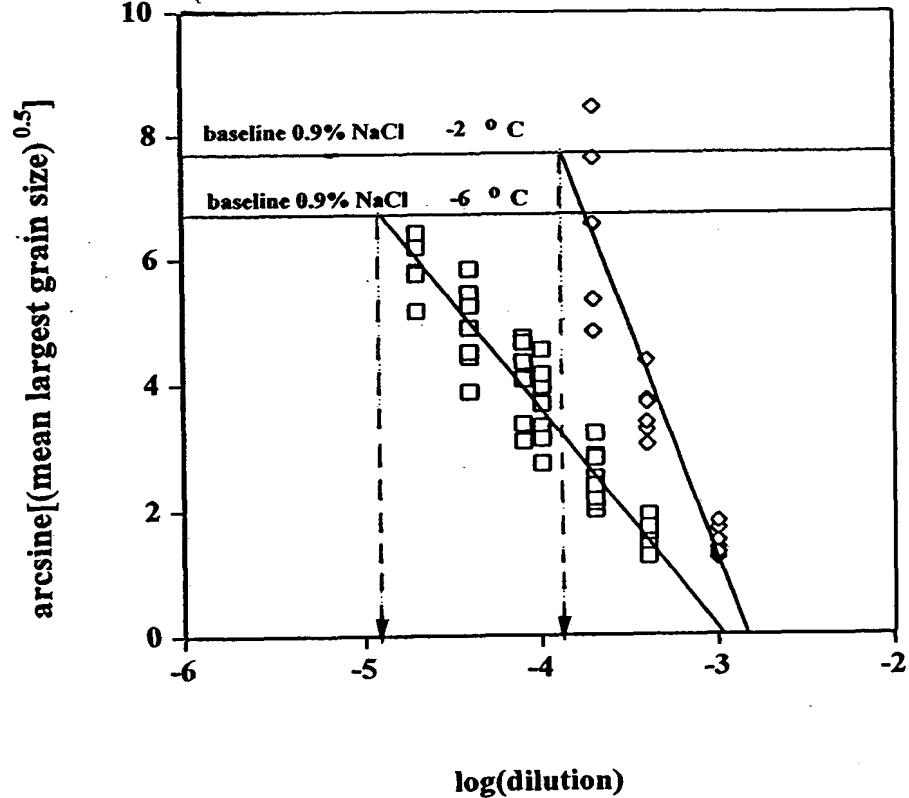


Fig. 8.20

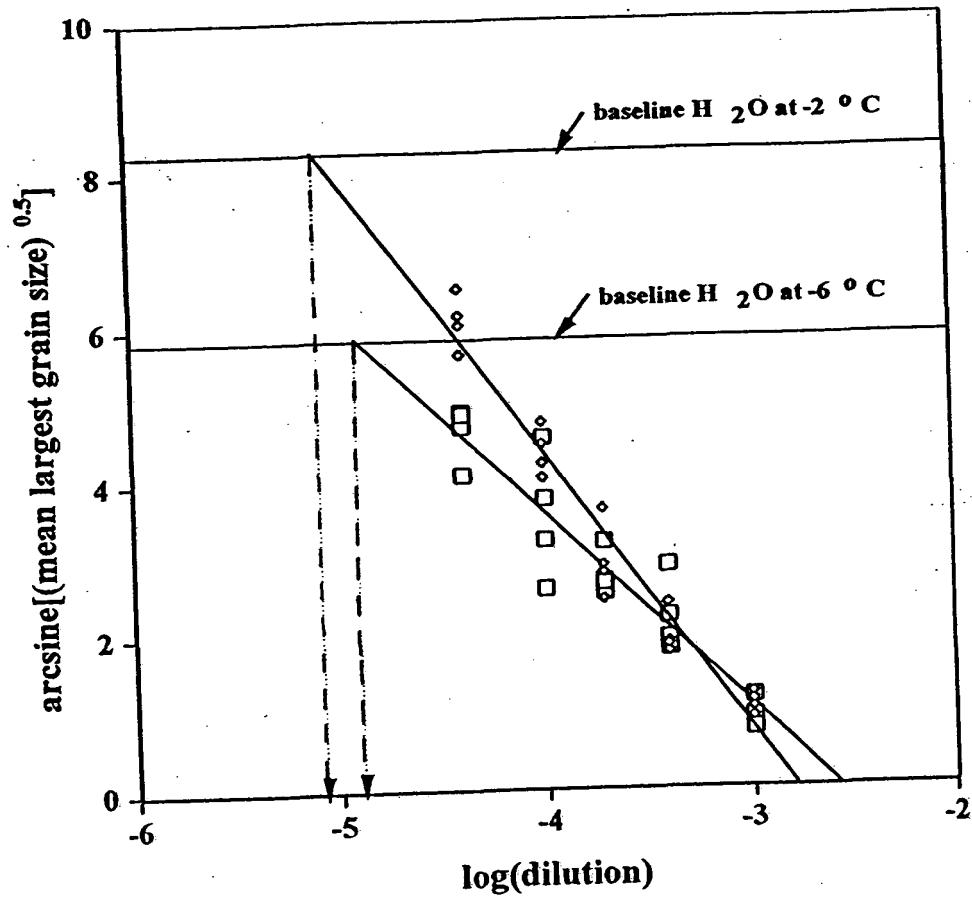


Fig. 8.21

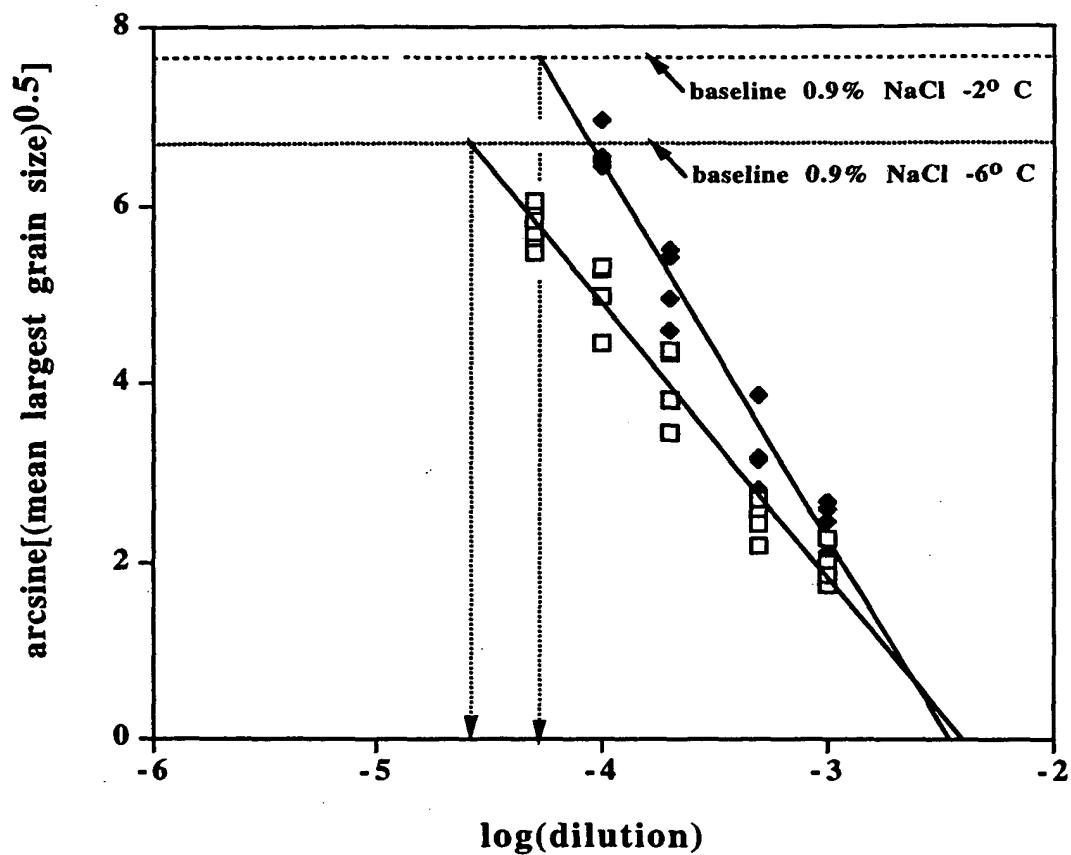


Fig. 8.22

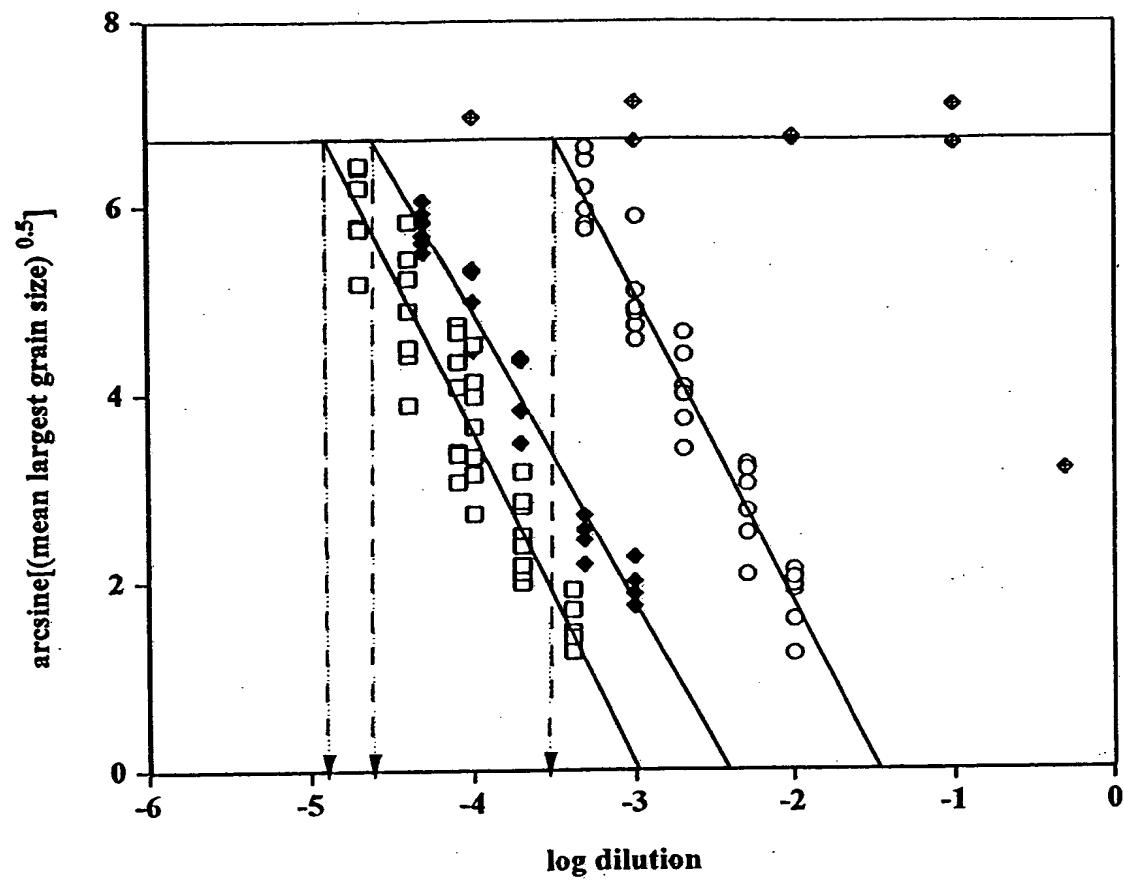


Fig. 8.23

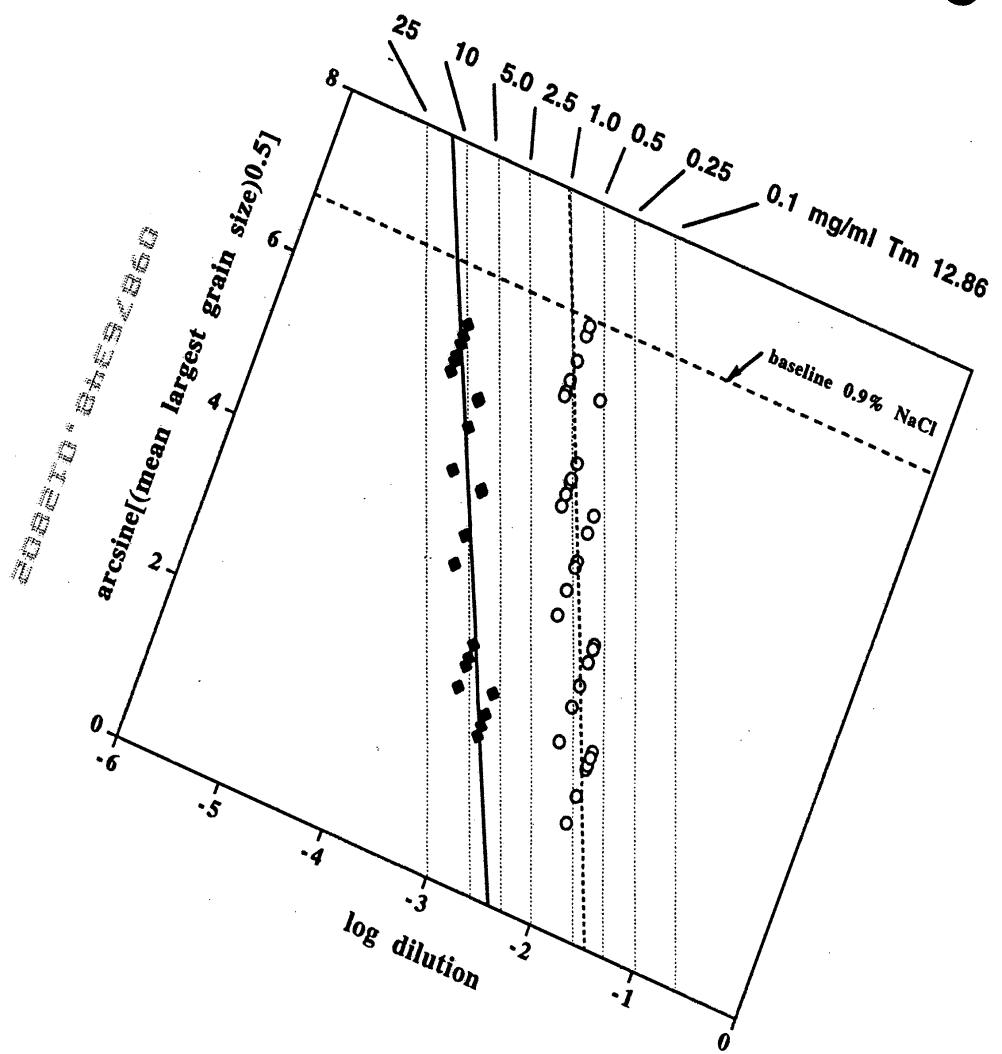


Fig. 8.24

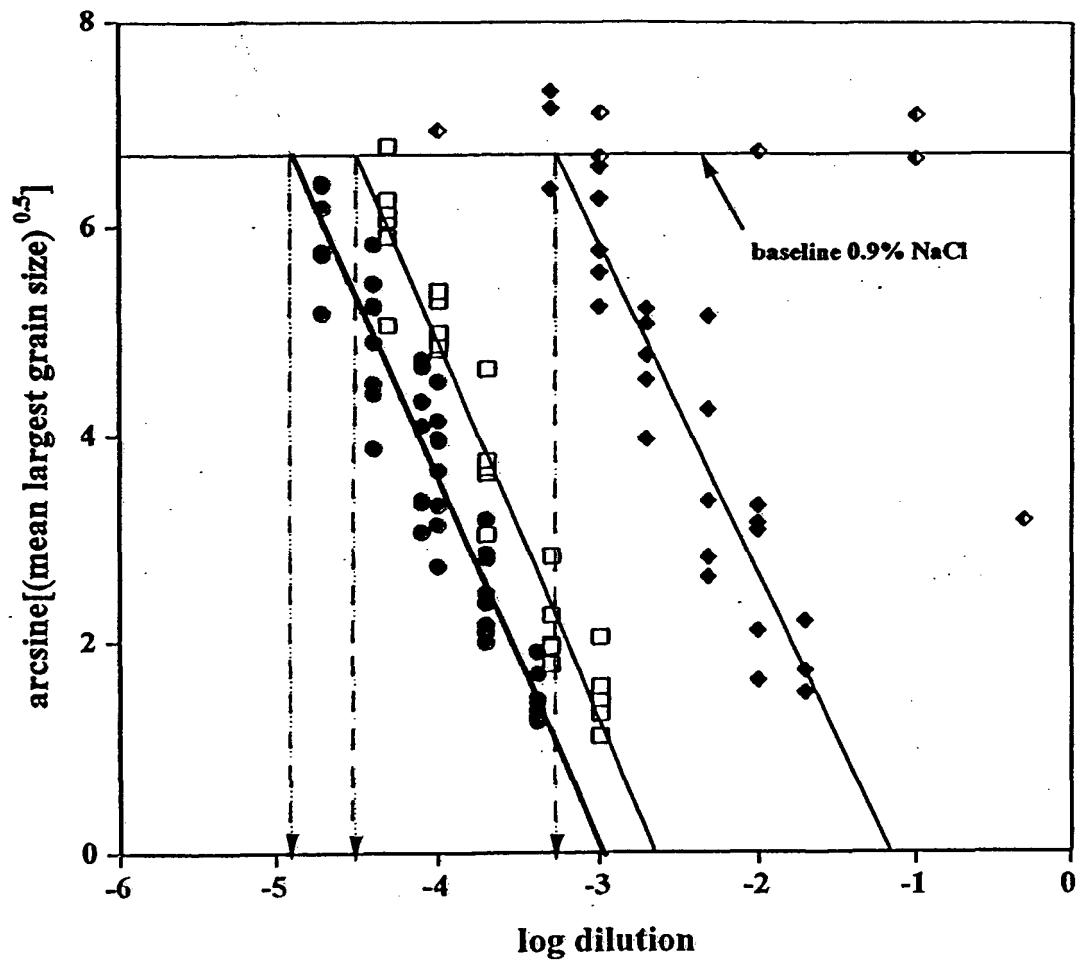


Fig. 8.25

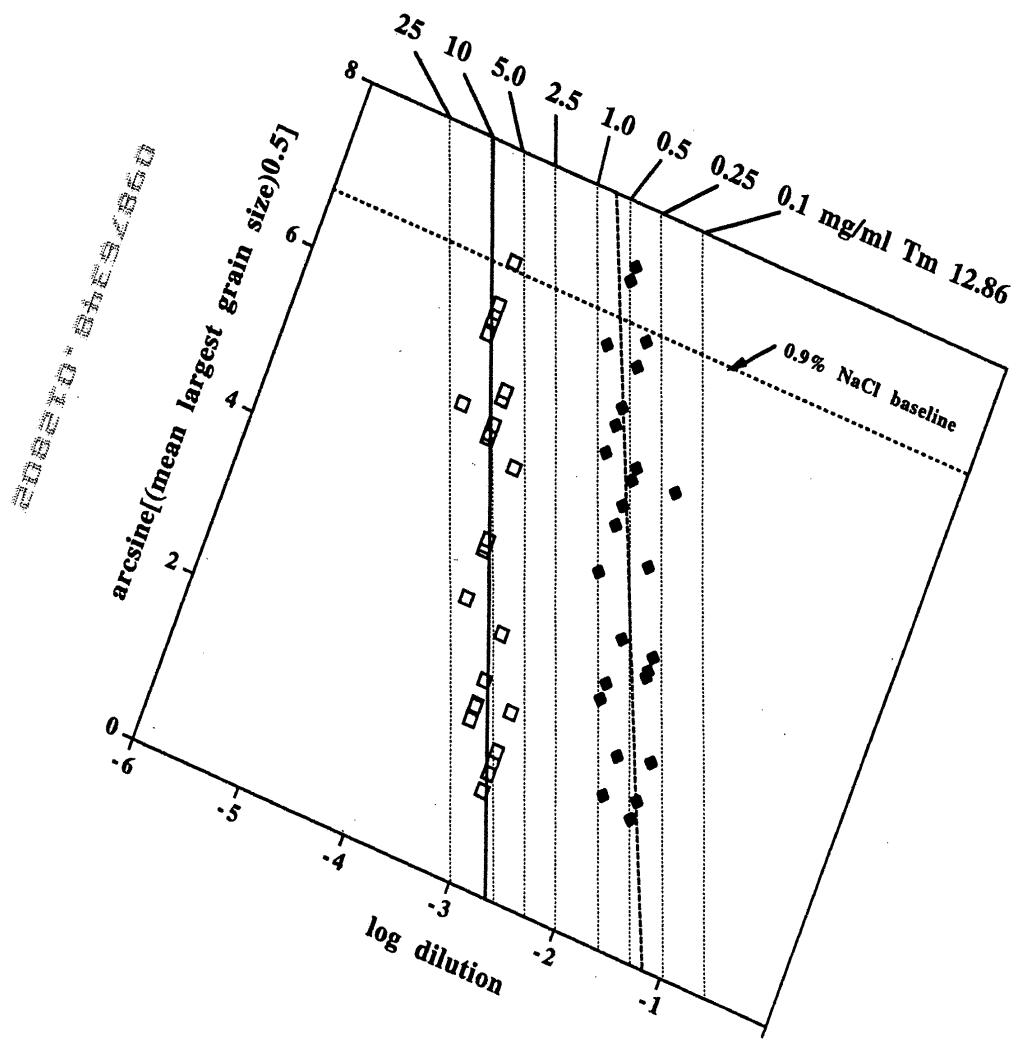


Fig. 8.26

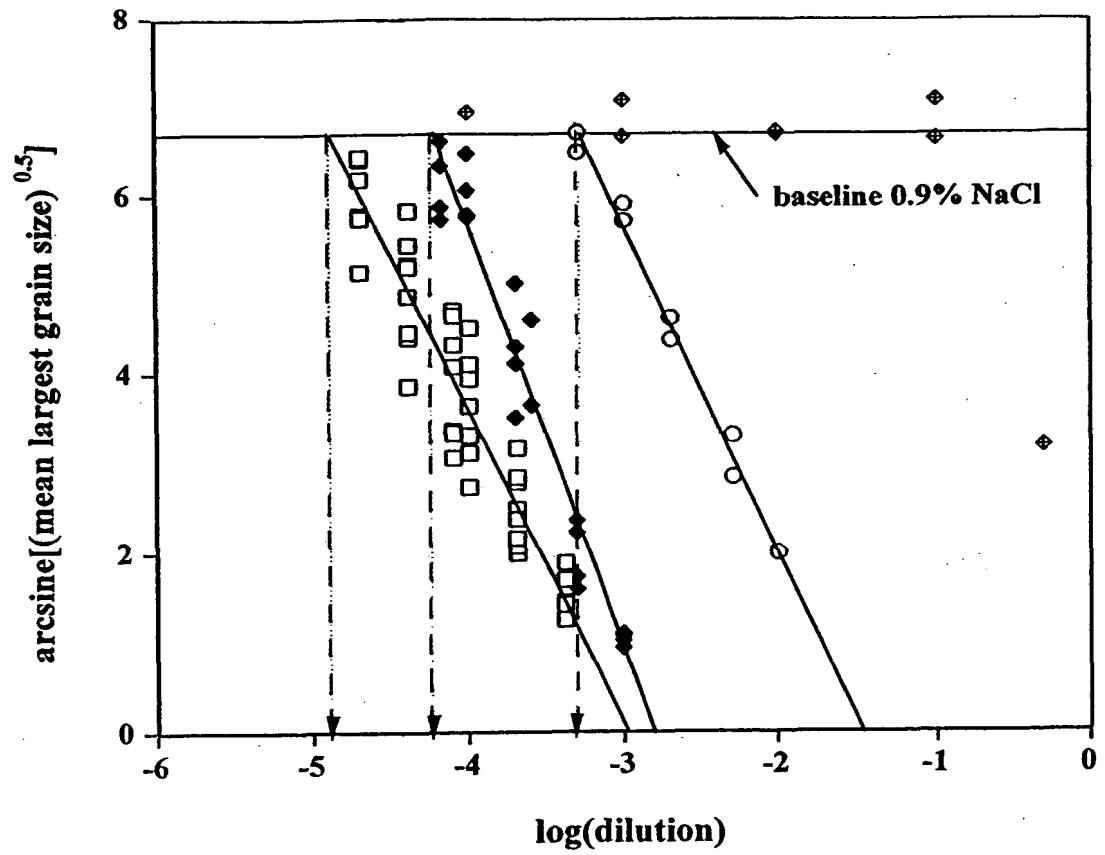


Fig. 8.27

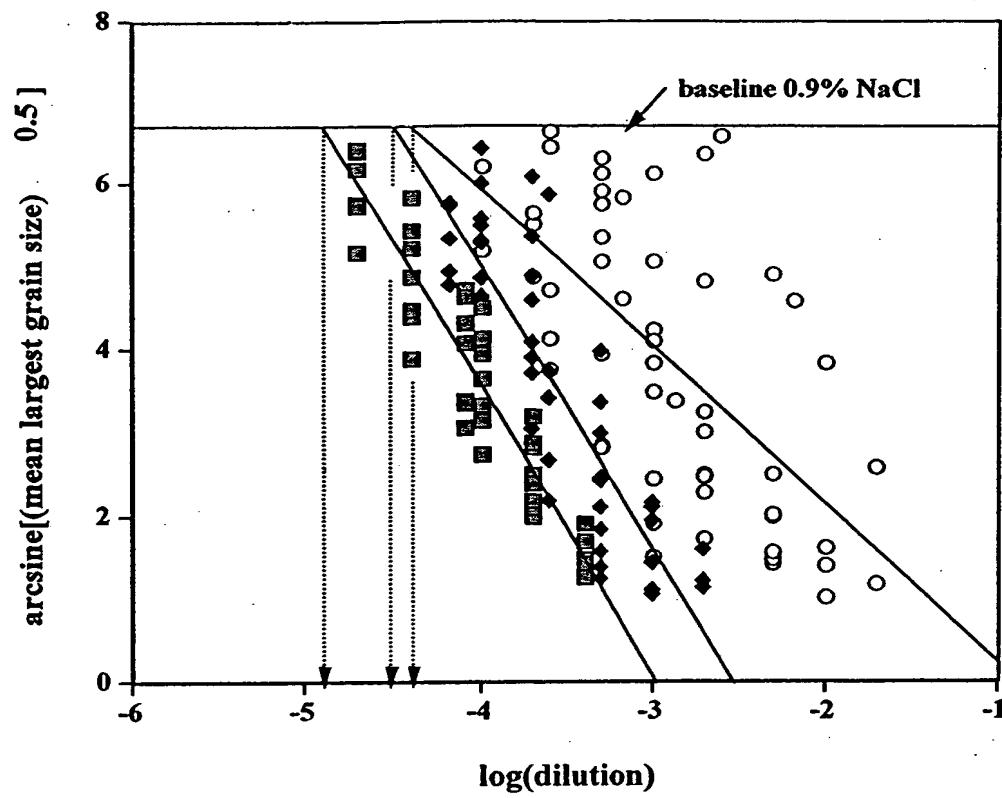


Fig. 8.28

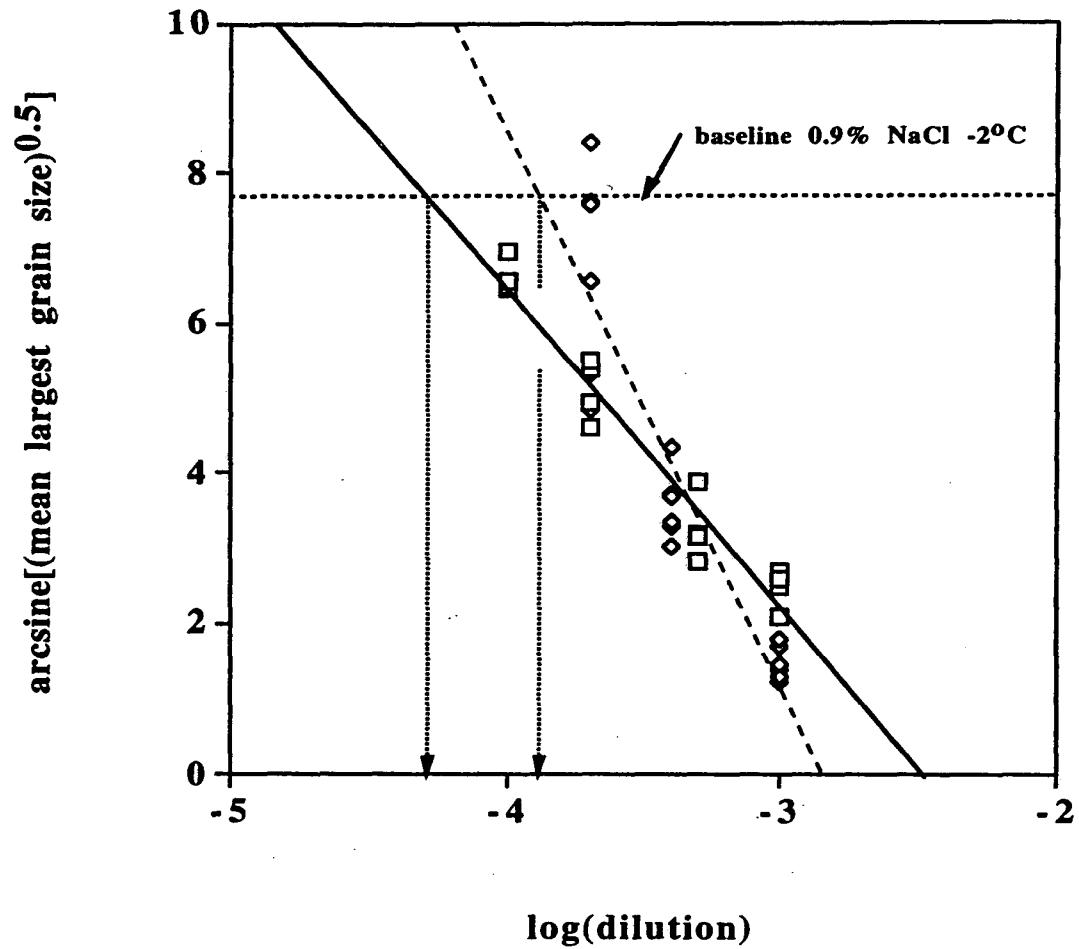


Fig. 8.29

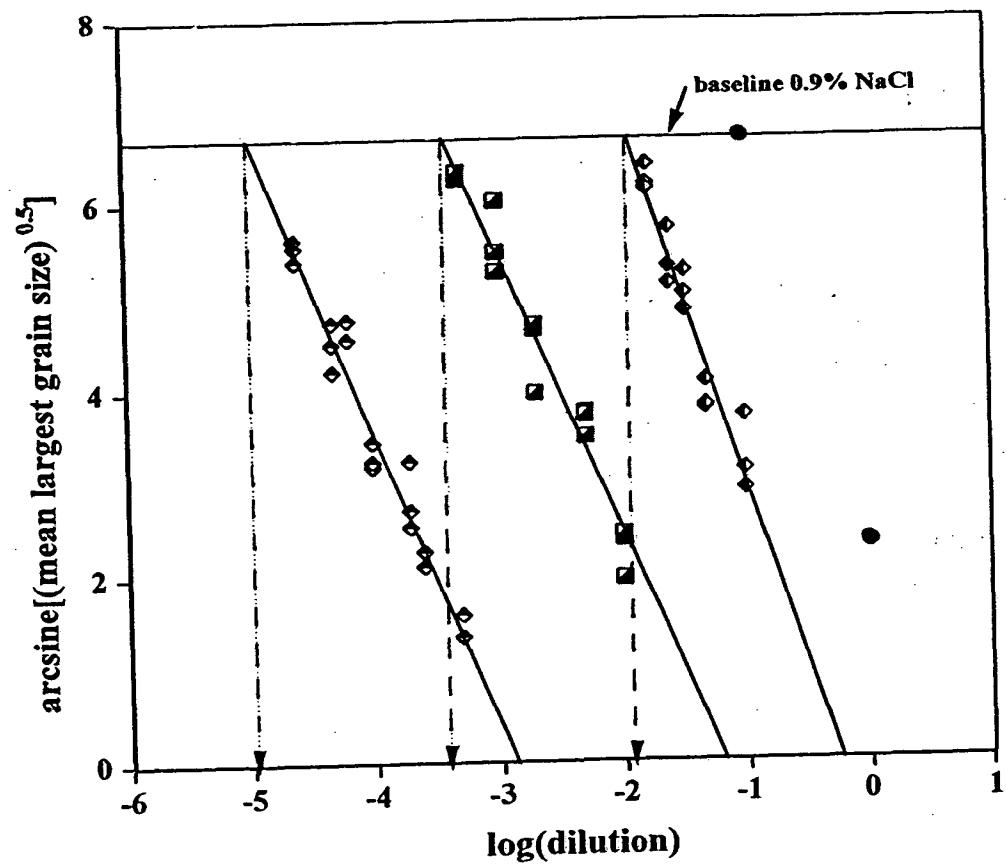


Fig. 8.30

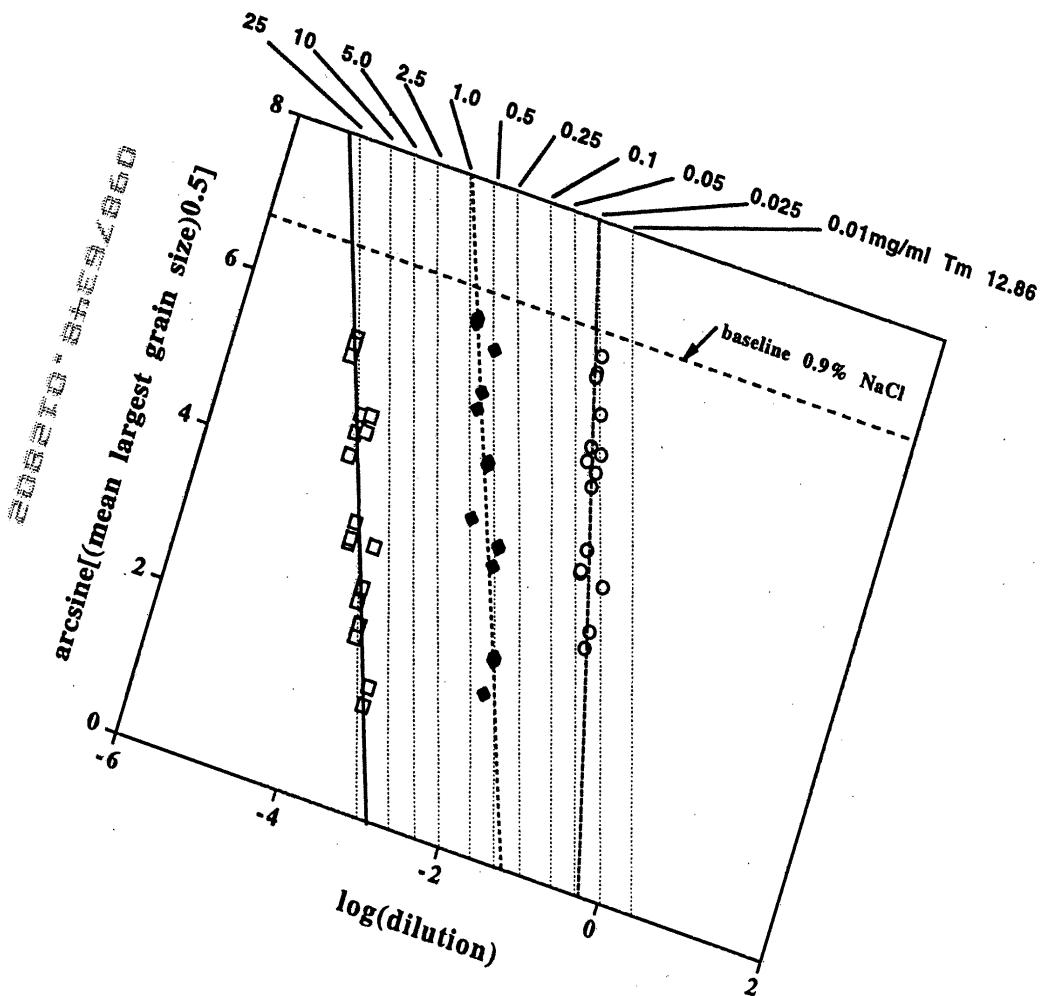


Fig. 8.31

mean largest grain size (mm^2)

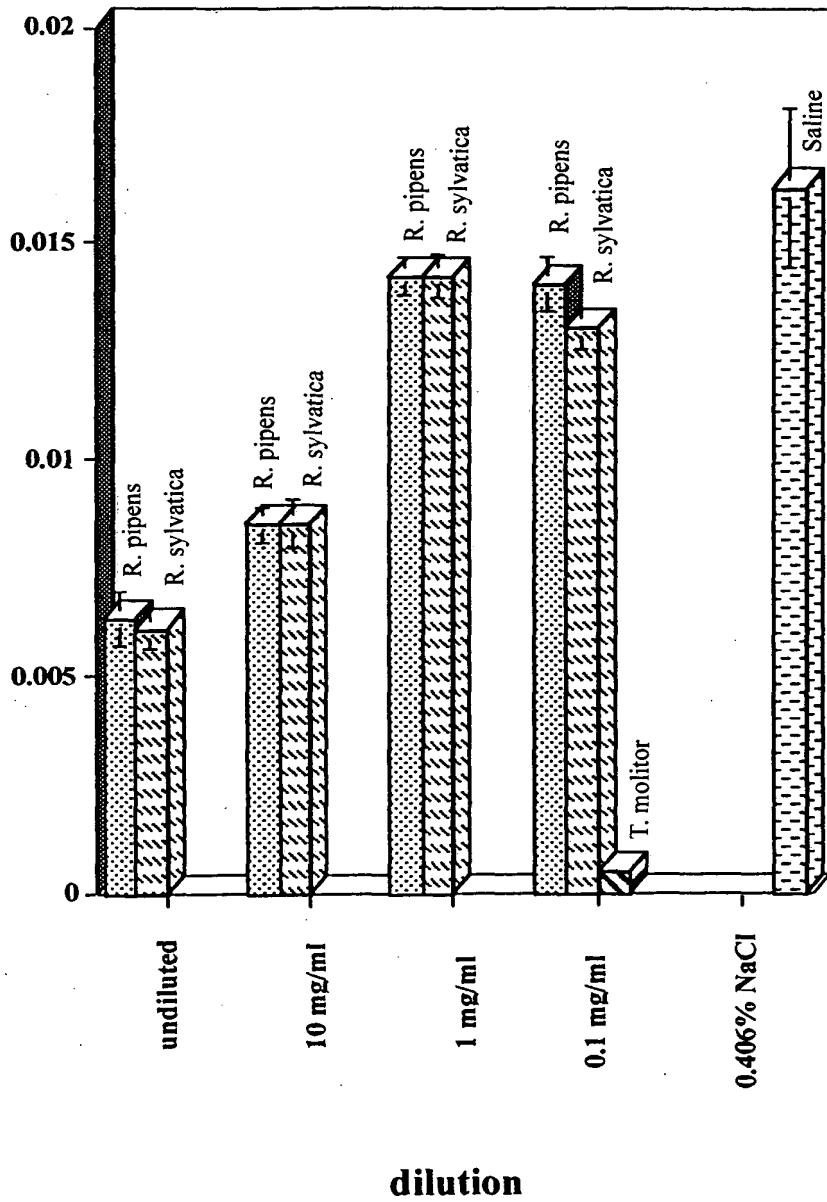


Fig. 8.32

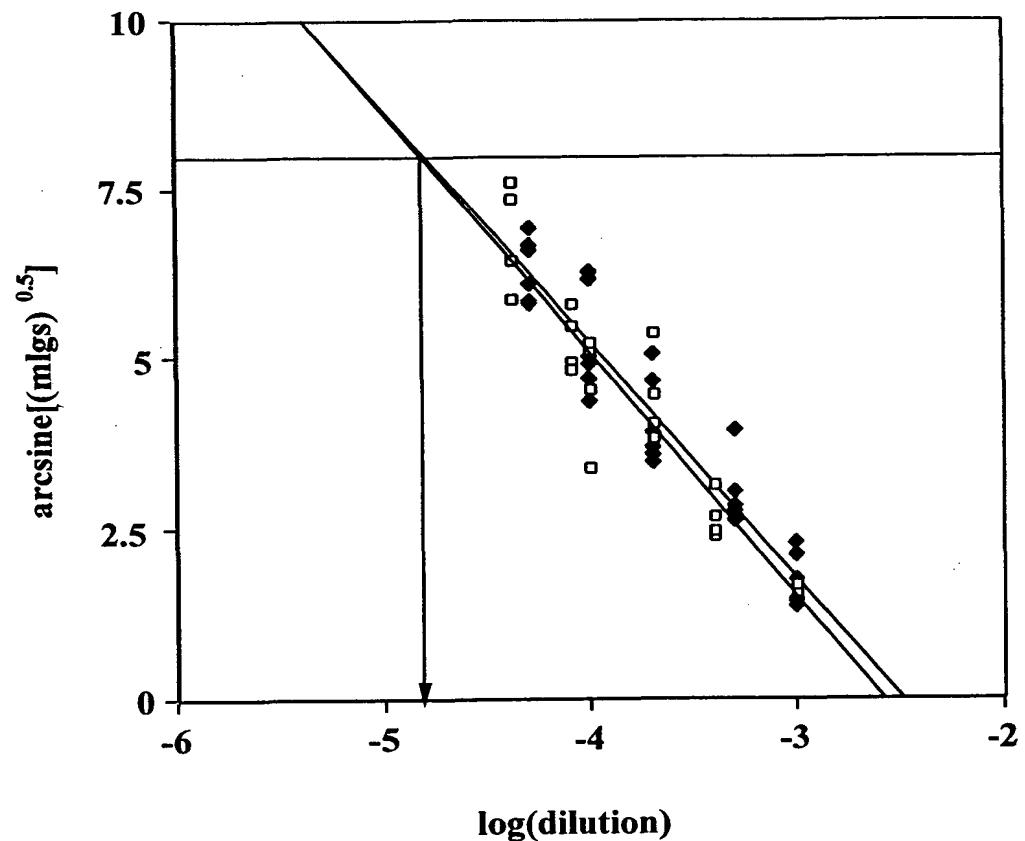


Fig. 8.33

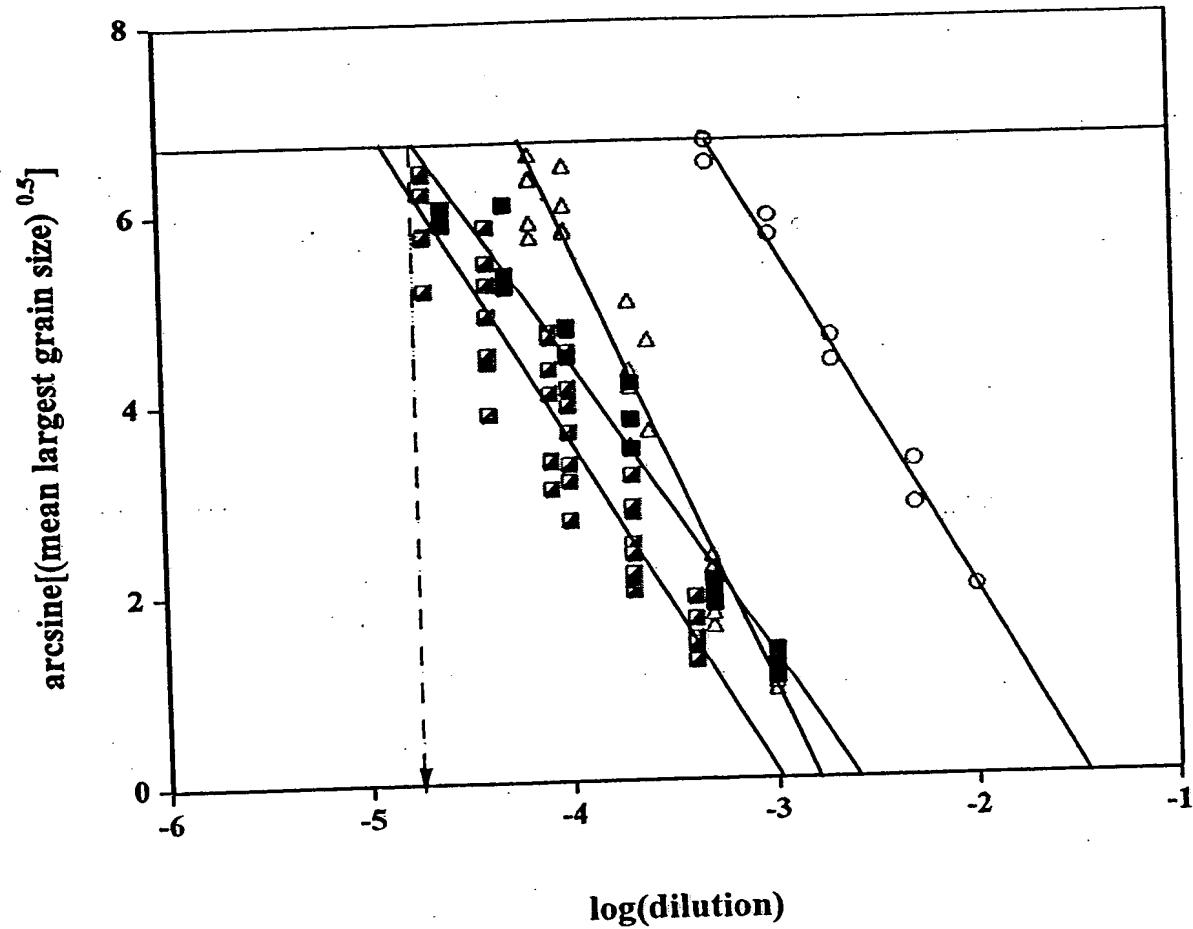


Fig. 8.34

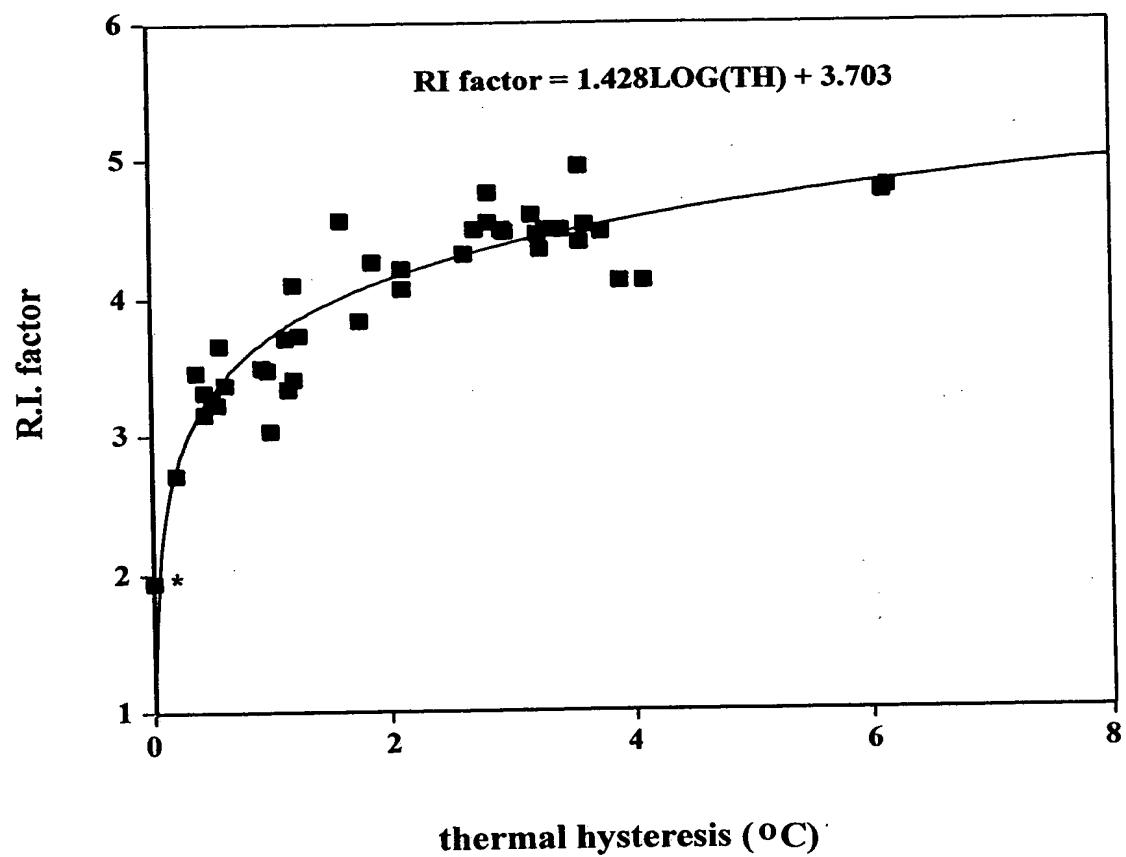


Fig. 8.35

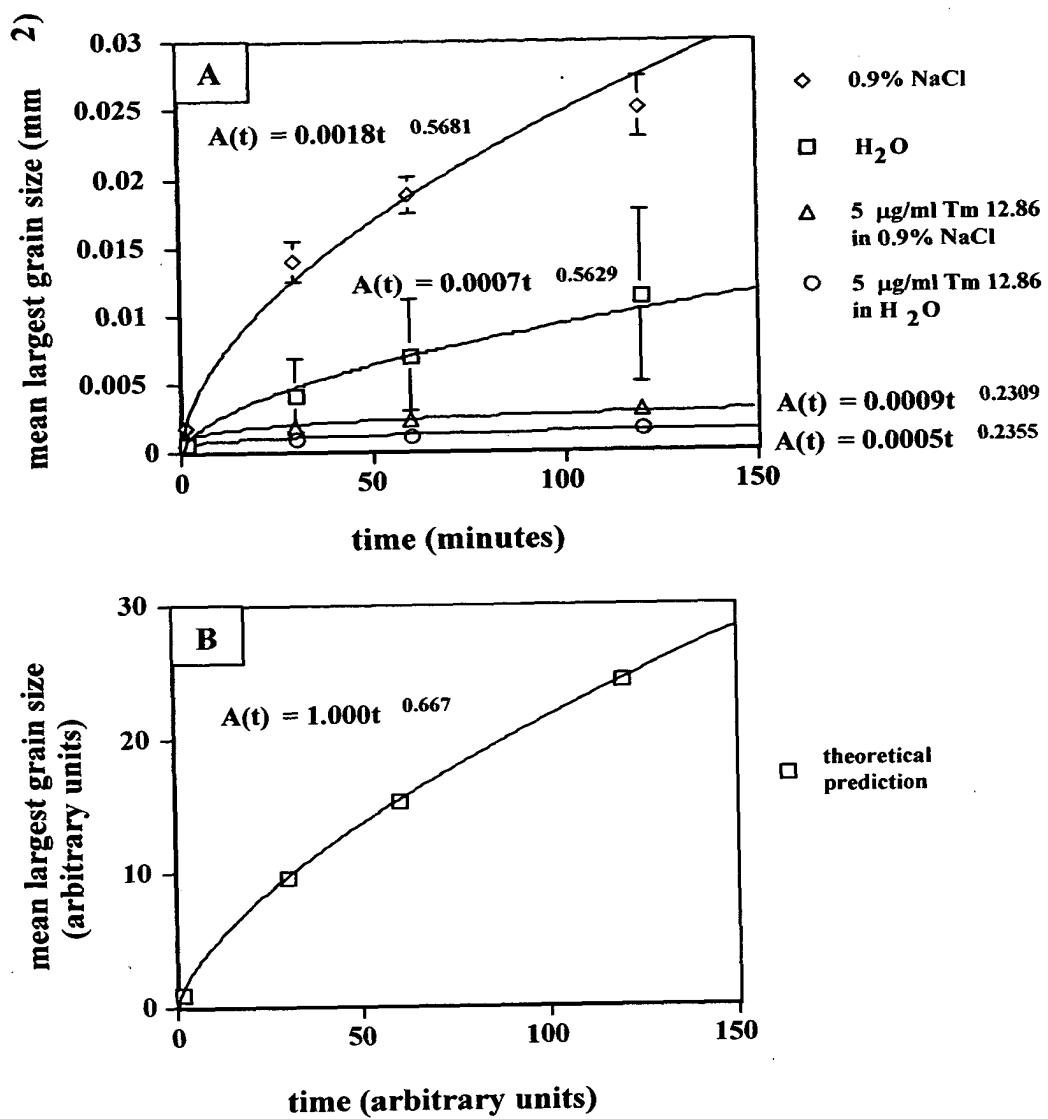


Fig. 8.36

2 1 3 2 1 3 2 1 3 2 1 3 2 1

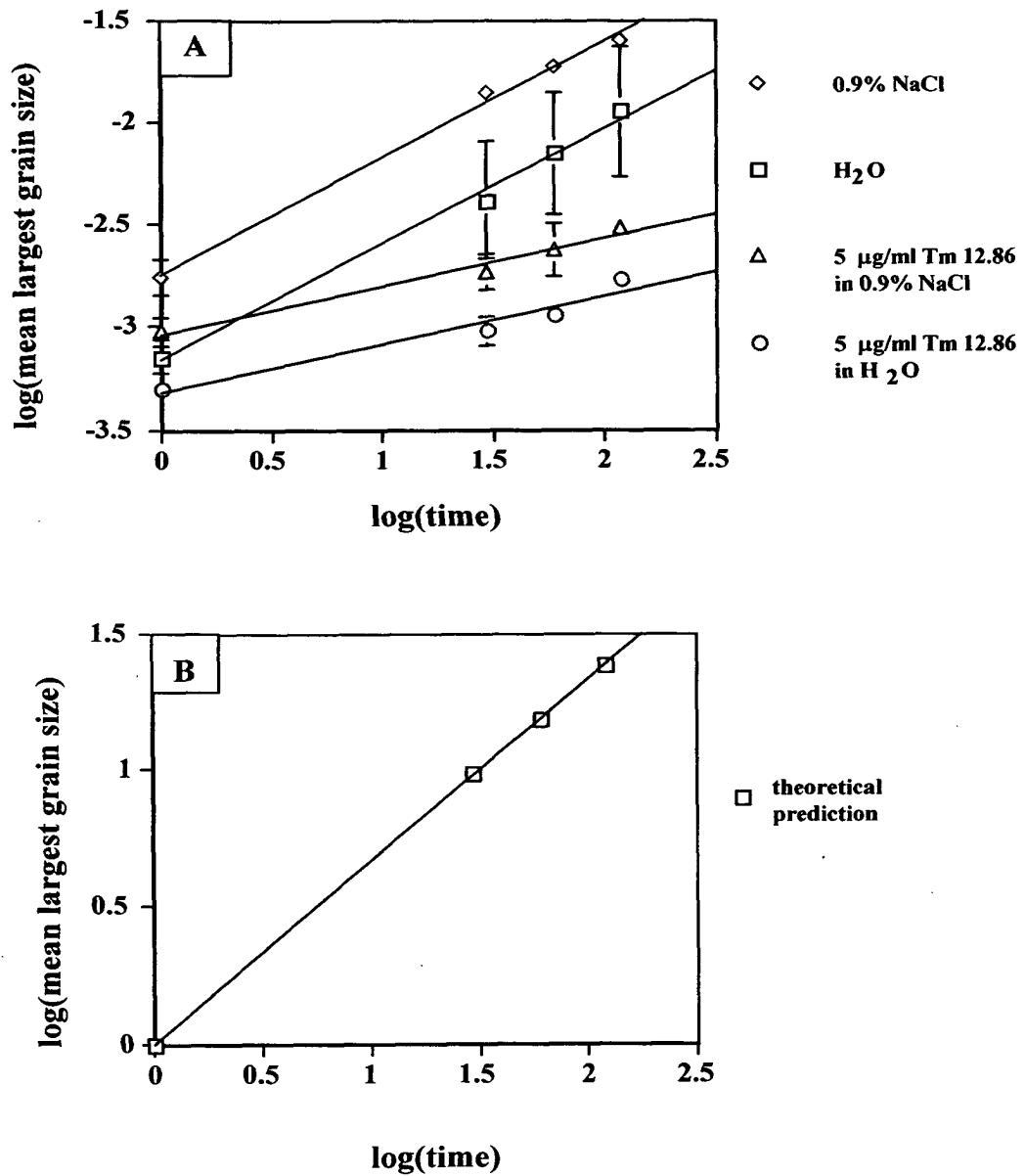
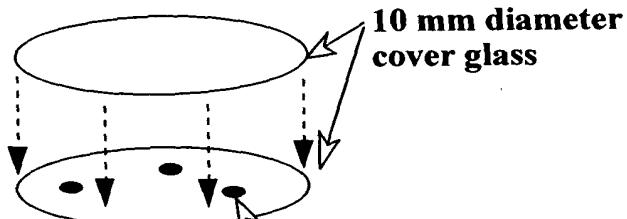


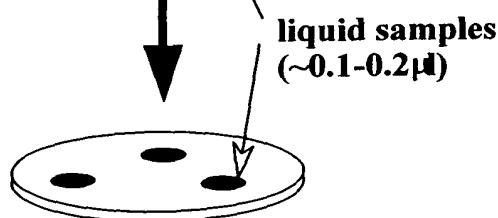
Fig. 8.37

"Sandwich" method of R.I. assessment

1.



2.



3. FREEZE ON ~80 C
ALUMINUM PLATE (~10 MIN.)



4. PLACE ON COLD STAGE,
ANNEAL AT -6 C UP TO
12+ HOURS

Fig. 8.38

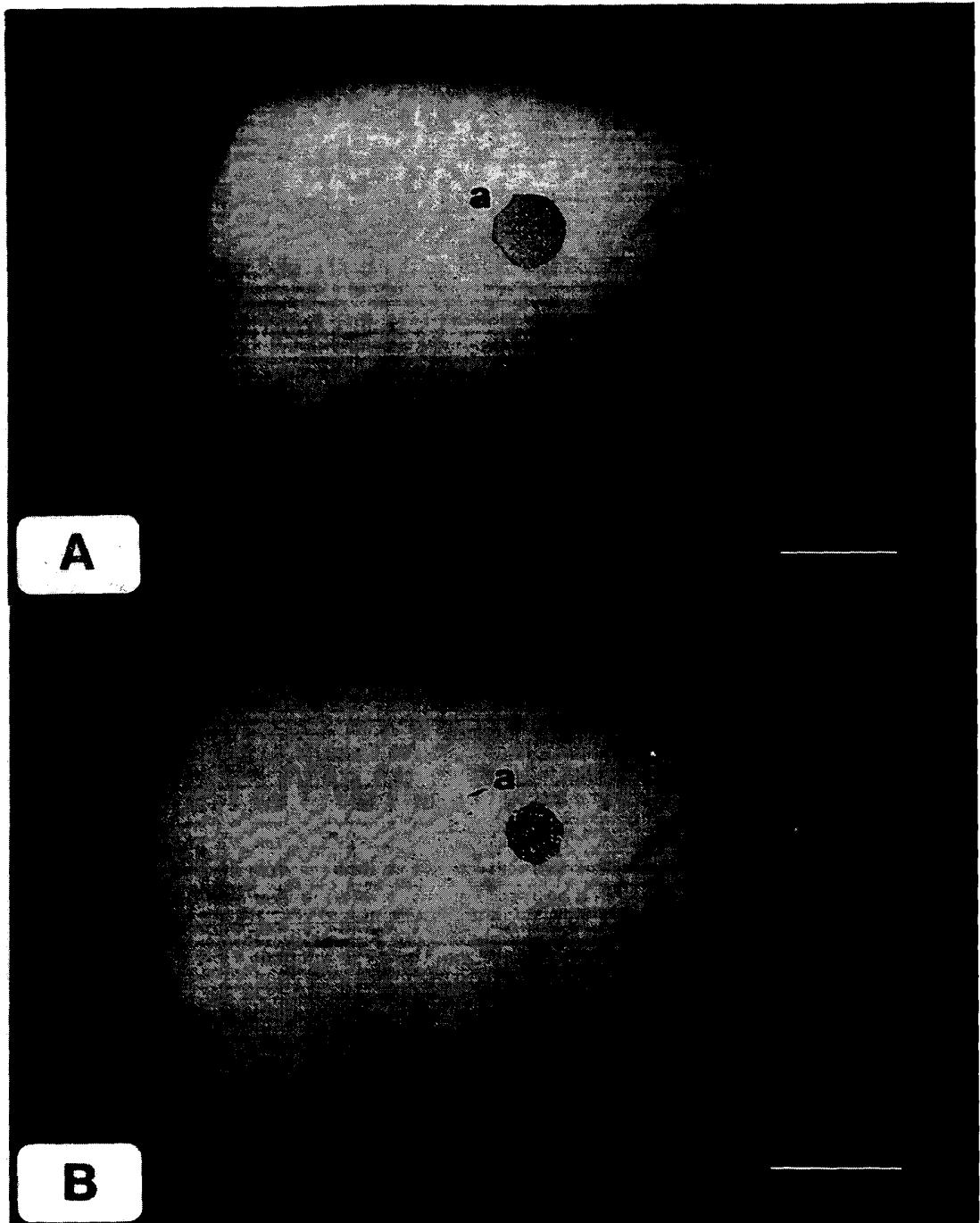


Fig. 8.39

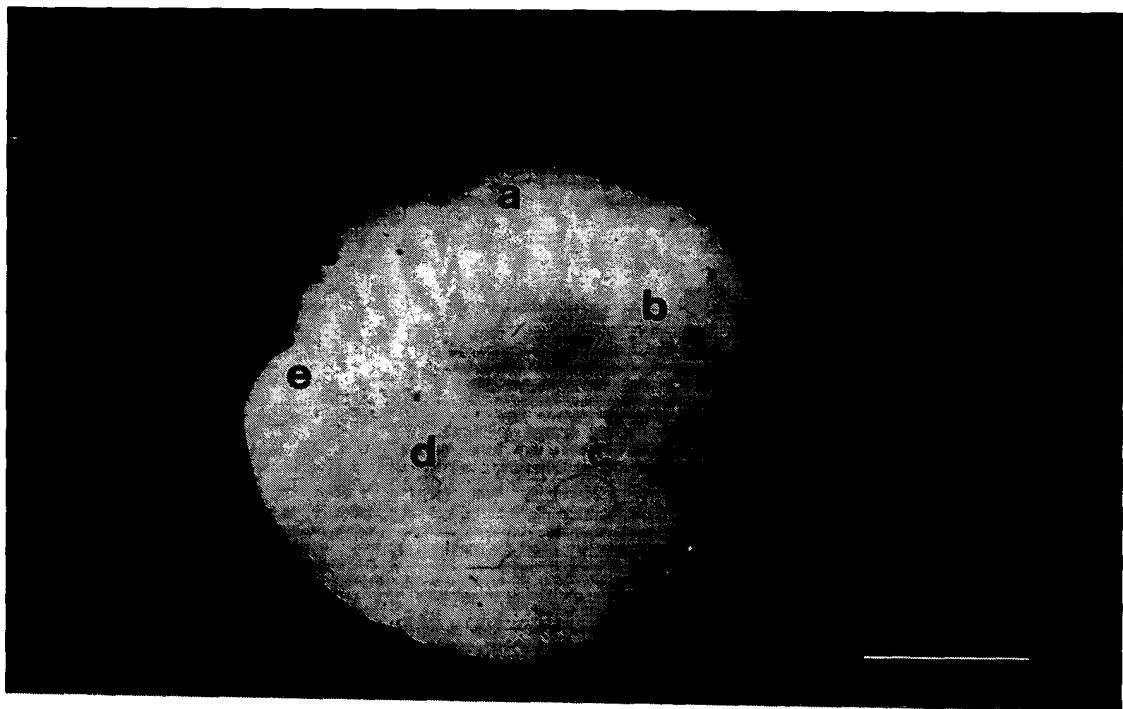


Fig. 8.40

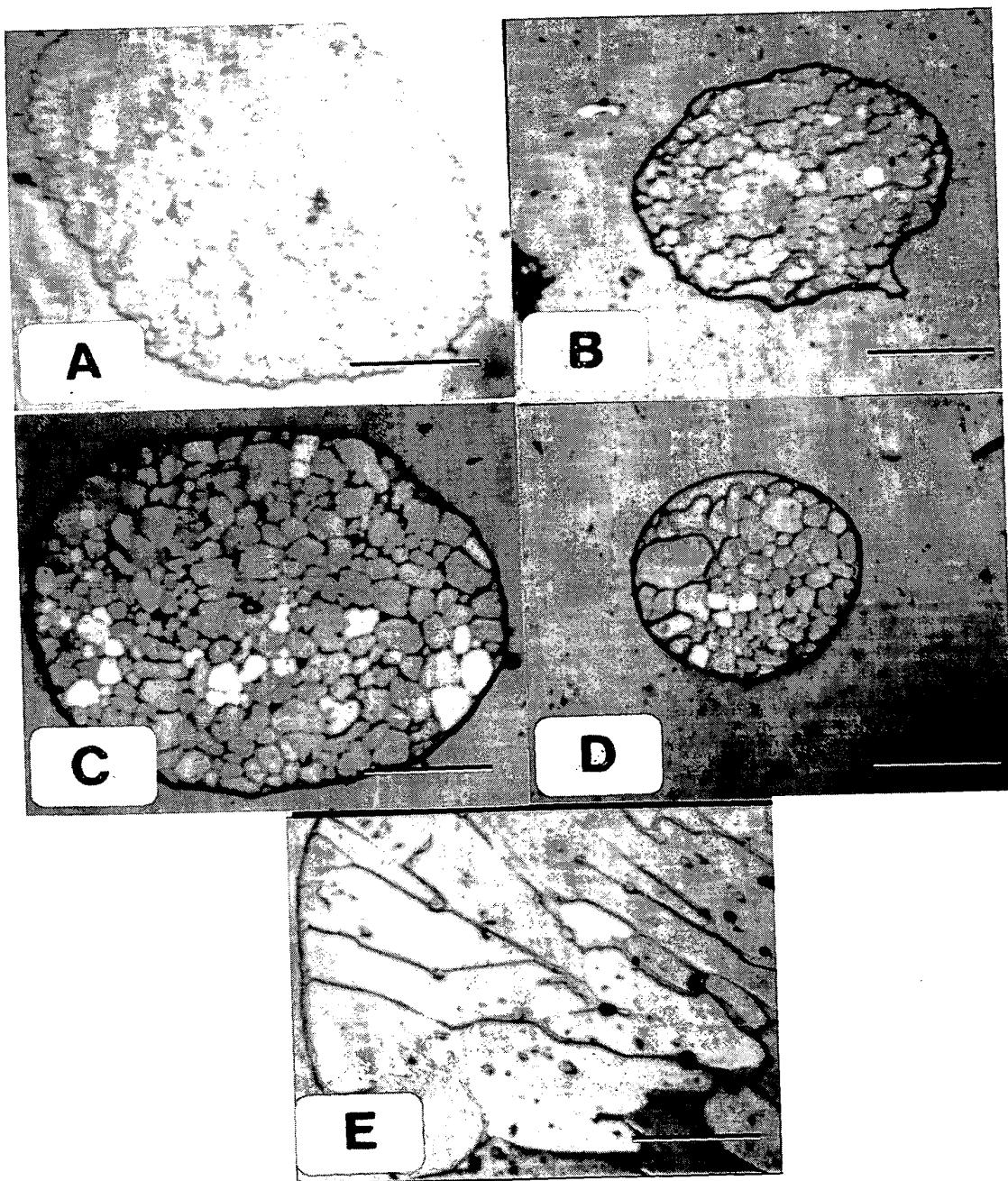


Fig. 8.41

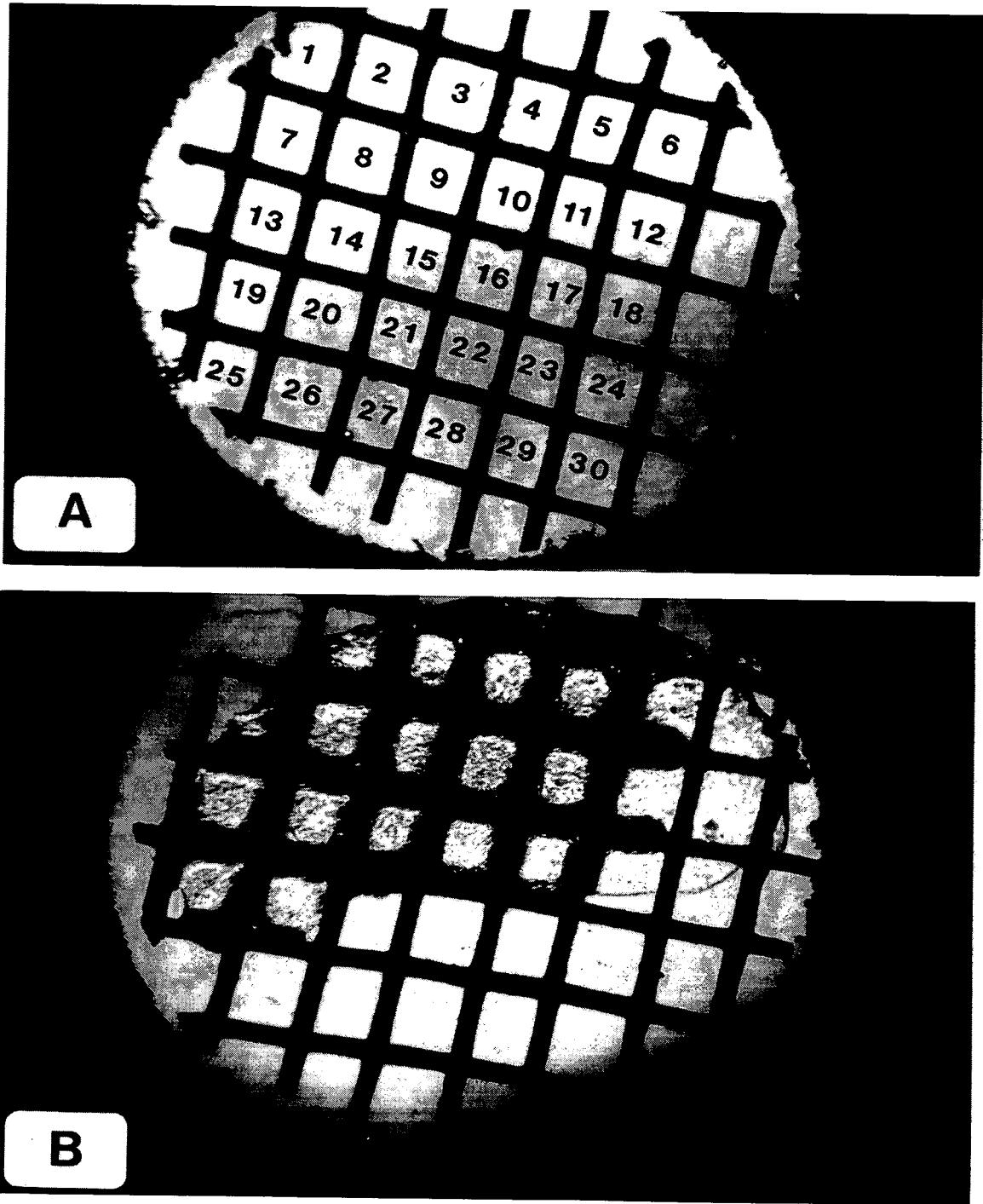


Fig. 8.42

DNA sequence of Tm 13.17 cDNA clone

B	E
a	C
m	O
H	R
I	I

1	AGTGGATCCAAGAATTGGGACGGAGACTACTAAAG	TGAAGTTGCTCTGTTGTCTAATCT	M K L L C C L I S
61	CCCTCATTCTGTTGGTCACAGTTCAAGGCC	TGACCGAGGCACAAATTGAGAAACTGAACA	G P
	L I L L V T V Q A	L T E A Q I E X K L N K	
		↑	
121	AGATCAGCAAAAAATGTCAAAATGAAAGTGGAGTGTCGCAAGA	GATCATACCAAGCTC	
	I S K K C Q N E S G V S Q E I I T K A R		
181	GCAACGGTGACTGGGAGGACGATCCTAAACTGAAACGCCAAGTTTTGCGTGGCCAGGA	N G D W E D D P K L K R Q V F C V A R N	
241	ACGCCGGTCTGGCCACGGAATCGGGAGAGGTGGTGGTCGACGTGTTGAGGGAGAAGGTGA	A G L A T E S G E V V V D V L R E K V R	
301	GGAAGGTCACTGACAACGACGAAGAAAATGAGAAAATCATCAATAAGTGCAGCCGTCAAGA	K V T D N D E E T E K I I N K C A V K R	P
361	GAGATACTGTTGAAGAGACGGTGGTCAATACTTTCAAATGTGTCAATGAAAAACAAGCCAA	D T V E E T V F N T F K C V M K N K P K	
421	AGTTCACCAAGTTGATTGAAACCACCAACGACTAGTAGATGGTTCAAATGGTGTGCTTAC	F S P V D *	
			X
			h
			o
			i
481	ATATAAAAATAAAAGTGTCTGATGTA	AAAAAAAAAAAAAAAAAAACTCG	
	polyadenylation signal poly (A) tail (26)		
537	AGAGTATTCTAGAGCGGCCGCGGCCATCGTTCCACCC		

Fig. 8.43

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P

1 GGCACGAGCAAAATGAAACTCCTCTTGTGCTTGCGTCGCC
M K L L L C F A F A A

P G

47 ATCGTCATCGGAGCTCAGGCTCTCACCGACGAACAGATAACAGAAA
I V I G A Q A L T D E Q I Q K

Y Y

92 AGGAACAAGATCAGCAAAGAACATGCCAGCAGGTGTCCGGAGTGTCC
R N K I S K E C Q Q V S G V S

Y

137 CAAGAGACGATCGACAAAGTCCGCACAGGTGTCTTGGTCATGAT
Q E T I D K V R T G V L V D D

182 CCCAAAATGAAGAACGACGTCCCTCTGCTTCTCGAAGAAAACTGGA
P K M K K H V L C F S K K T G

226 GTGGCAACCGAACGCCGGAGACACCAATGTGGAGGTACTCAAAGCC
V A T E A G D T N V E V L K A

271 AAGCTGAAGCATGTGGCCAGCGACGAAGAGGTGGACAAGATCGTG
K L K H V A S D E E V D K I V

P

316 CAGAAGTGCGTGGTCAAGAACGGCCACACCAAGAGAAACGGCTTAT
Q K C V V K K A T P E E T A Y

P G

361 GACACCTTCAAGTGTATTTACGACAGCAAACCTGATTCTCTCCT
D T F K C I Y D S K P D F S P

G

406 ATTGATTAATTGTTTGTATTTGACTGAATTTGACAATAAGGT

polyadenylation signal

451 ACTATCGTTATGTAAAAAAAAAAAAAA

poly (A) tail

Fig. 8.44